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Formerly Cell Biosciences

FluorChem E System User Guide

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FluorChem E User Guide

P/N 94-15038-00

Revision 4a, June 2012

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Chapter 1:

General Information

Chapter Overview

- Welcome
- Introduction to the FluorChem E System
- Safety
- Customer Service and Technical Support
- Legal Notices

Welcome

Congratulations on your purchase of the FluorChem® E system. ProteinSimple welcomes you as a new user. This user guide includes important safety information, installation requirements, maintenance procedures, descriptions of the instrument and user interface as well as detailed operating instructions for setting up and running FluorChem E.

Introduction to the FluorChem E System

FluorChem E is a high-performance imaging system for quantitative Western blot detection and gel imaging. Its compact design incorporates state-of-the-art CCD optical technology with an intuitive touchscreen interface for detection of chemiluminescent, UV-excited and colorimetric gels and blots. In addition, users can control and monitor image acquisition and access data remotely using a unique, browser-based interface from any networked computer or mobile Wi-Fi device.

The FluorChem E system comes with a cooled 8.3 megapixel CCD detector, 50-mm f/1.4 motorized fixed lens, 4-position motorized filter wheel and 590/50 nm orange emission filter. Light sources include epi-white lights, trans-white light via white light conversion screen and an integrated 302 nm UV transilluminator. The system also includes an on-board integrated computer and LCD touchscreen, the FluorChem E Digital Darkroom User Interface and AlphaView® stand alone image analysis software.

Safety

User Attention Notifications

Several user attention phrases are used throughout this manual. Each phrase should draw the following level of attention from the user:

NOTE Points out useful information.

IMPORTANT Indicates information necessary for proper instrument operation.

CAUTION Cautions users regarding potentially hazardous situations in regard to user injury or damage to the instrument if the information is not heeded.

!WARNING! Warns users that serious physical injury can result if warning precautions are not heeded.

Instrument Safety Labels

The following safety labels are located on the FluorChem E system. Each label will display a safety alert symbol indicating a potential safety hazard.

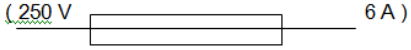
Symbol	Description
	Ultraviolet Light
	Risk of Electric Shock
	Refer to the FluorChem E User Guide before proceeding
	Hot Surface
	Fuse Symbol (6 A fuses only)

Table 1-1: *FluorChem E system safety labels.*

!WARNING!
If the system is not used as specified by ProteinSimple, overall safety will be impaired.

!WARNING!
If the system is damaged and does not function properly, stop the instrument safely and contact ProteinSimple Technical Support immediately.

UV Safety

CAUTION
The FluorChem E system contains an integrated ultraviolet transilluminator. The system enclosure confines the radiation within the system and shields the user from exposure. Exposure to UV radiation can cause permanent damage to the eyes and skin.

UV Interlock Switch

The FluorChem E system is equipped with a safety interlock switch which automatically cuts off the power to the transilluminator when the door is open. This eliminates any dangerous user exposure to UV light. As a safety precaution, the UV light will automatically turn off after three minutes when using the interlock switch.

To use the transilluminator for other purposes, a key is required to override the safety interlock. In this case, be sure to use the following safety precautions:

- Always wear UV-protected eyewear specified by the manufacturer as providing protection at the wavelength(s) used, making sure the eyewear protects any areas where radiation may come through. UV sunglasses may not prevent UV radiation from coming in through the sides or around the lenses.
- Always cover all skin that may be exposed to UV light, in particular the face, neck, hands and arms.
- Always make sure that any UV protection devices (such as the safety interlock switch on the transilluminator) are working properly. If not, discontinue use until the device(s) are properly repaired.
- Use only UV lamps in the transilluminator.

Hot Surface

CAUTION

Under normal conditions, the temperature of the glass surface of UV transilluminator is safe to touch. However, if the system malfunctions, it is possible that the glass surface temperature could exceed 80 °C. Please exercise caution when touching the glass surface.

Electrical Safety

Be sure to take proper precautions when handling any electrical equipment. Never work on any live circuit, fixture, receptacle, or switch. Safety rules to follow whenever working with any electrical appliance include:

- Always turn the system power off and unplug the power cord before changing a fuse.
- Always shut off power to the circuit before repairing or replacing a switch, receptacle, or fixture.
- Always tape over the main switch, empty fuse socket, or circuit breaker being worked on.
- Always check that the circuit is dead before beginning work on it. Using a circuit tester or voltmeter can help determine this.
- Always unplug any appliance before repairing it.

Prior to System Operation

Ensure that all users of the FluorChem E system have:

- Received instruction in general safety practices for laboratories.
- Received instruction in specific safety practices for the instrument.
- Received instruction on handling of biohazards if biohazardous materials are to be used on the system.

CAUTION

Avoid using the FluorChem E system in a manner not specified by ProteinSimple. While the system has been designed to protect the user, this protection may be impaired if the instrument is used improperly.

Customer Service and Technical Support

The ProteinSimple support staff is available to respond to any questions or comments about our products. For questions or general feedback, use the following contact methods:

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Legal Notices

NOTE: Read the Legal Notices carefully before using your FluorChem E system.

FluorChem E System Disclaimer of Warranty

EXCEPT AS EXPRESSLY PROVIDED IN ANY ProteinSimple SOFTWARE LICENSE AGREEMENT OR QUOTATION, THE PRODUCTS SOLD AND SERVICES PROVIDED BY ProteinSimple ARE PROVIDED ON AN "AS IS" AND "AS AVAILABLE" BASIS WITHOUT WARRANTY OF ANY KIND. ProteinSimple AND ITS SUPPLIERS DO NOT WARRANT THE SECURITY, PRIVACY, OR ACCURACY OF ANY DATA PROVIDED VIA THE PRODUCTS OR SERVICES, AND YOU AGREE THAT THE USE OF ANY SUCH DATA BY YOU IS AT YOUR SOLE RISK. TO THE MAXIMUM EXTENT ALLOWED UNDER APPLICABLE LAW, ProteinSimple AND ITS SUPPLIERS DISCLAIM ANY AND ALL WARRANTIES, WHETHER EXPRESS, IMPLIED OR STATUTORY, INCLUDING, WITHOUT LIMITATION, ANY WARRANTY OF MERCHANTABILITY, TITLE, NON-INFRINGEMENT, OR FITNESS FOR A PARTICULAR PURPOSE.

Chapter 2:

Installation

Chapter Overview

- Introduction
- Space Requirements
- Electrical Requirements
- Environmental Requirements
- Software and Computer Requirements
- System Installation

Introduction

This chapter will help prepare the laboratory for installation of the ProteinSimple FluorChem E system. Please complete the space, electrical and environmental requirements prior to installing the instrument.

Space Requirements

The FluorChem E system is a bench-top instrument which requires a stable laboratory bench or table capable of supporting 75 lbs (34 kg).

System dimensions and space requirements are as follows:

Dimension	Centimeters	Inches
Width	32	12.6
Depth	62	24.5
Height	62	24.5

***Table 2-1:** FluorChem E system dimensions.*

A minimum clearance of 3 inches (7.6 cm) at the rear of the instrument must be provided for the CCD cooling fan ventilation and sufficient access to connect or disconnect the power cord as well as to operate the power switch.

Electrical Requirements

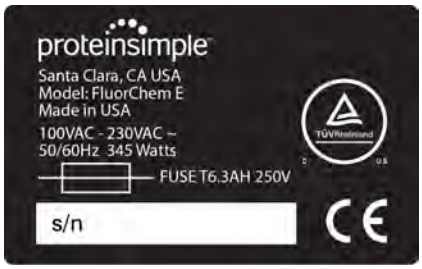
As with all electrical instruments, the FluorChem E system should be located away from water, solvents or corrosive materials. The system should also be placed away from interfering electrical signals and magnetic fields.

Installation of the FluorChem E system requires a dedicated, grounded circuit capable of delivering the appropriate current and voltage for your country. The power requirements for select countries are listed in Table 2-2.

Region	Volts (AC)	Frequency (Hz)	Amps
US and Canada	120	60	3
Europe	230	50	1.5
UK	230	50	1.5
Japan	100	50/60	3.5
China	220	50	1.6
India	230	50	1.5
South Korea	220	60	1.6

Table 2-2: *FluorChem E system power requirements.*

The FluorChem E system label displays the system power specifications:



In addition to the electrical power requirements listed above, ProteinSimple requires that all grounded circuits terminate at the receptacles, and the receptacles must be located within 10 ft (3 m) of the instrument.

Environmental Requirements

Please provide appropriate heating and cooling to maintain a constant laboratory temperature. For optimal FluorChem E system performance, the laboratory environment must meet the following criteria:

Requirement	Specification
Operating temperature range	15 - 25 °C (59 - 77 °F)
Operating humidity range	15 - 70% relative, non-condensing

***Table 2-3:** FluorChem E system environmental requirements.*

Software and Computer Requirements

The FluorChem E system includes an integrated computer with the FluorChem E User Interface pre-installed. Post-acquisition image analysis can be performed using AlphaView stand alone software.

Please use the AlphaView software CD provided with the FluorChem E system to install the software on a separate computer workstation, such as a user's desktop computer. In order to run AlphaView software, this computer must meet the minimum requirements listed in the table below:

Component	Minimum Recommended
Operating System	Windows 7, Vista or XP
Processor	Pentium 4 at 2.8 GHz
Memory	1 GB
Hard Drive	40 GB

***Table 2-4:** AlphaView software computer requirements when used on local workstations.*

Networking the FluorChem E System

The FluorChem E system can be connected directly to a local network for remote access using a web browser on any Mac or PC-based network computer or Wi-Fi enabled device such as a smartphone or iPad™. Image acquisition can be controlled and monitored remotely, and stored image files can be accessed for copying, printing and post-acquisition desktop analysis using AlphaView software. For more information on accessing the FluorChem E system from a remote computer or device, please refer to Chapter 9, *"Networking and Remote Access"*.

System Installation

The FluorChem E system is easily installed by the user. Once the space, electrical and environmental requirements have been met:

1. Unbox the system and place it on a lab bench.

IMPORTANT

The FluorChem E system weighs 75 lbs (34 kg). Two people are required to lift the system from the shipping container onto the laboratory bench. Lift the system by grasping the bottom of the instrument at the midsection on either side.

---

2. Connect the power cord to the rear panel of the system and plug the cord into a power receptacle.
3. Network access (optional). To access FluorChem E via a local network, connect an Ethernet LAN cable to the 10/100/BASE-T (RJ-45 connector) port.
4. Turn FluorChem E on using the power switch on the rear panel.
5. Let FluorChem E equilibrate for 30 minutes after the initial power up before imaging samples. This allows the system optics to reach the optimum acquisition temperature of -25 °C.

Chapter 3:

System Hardware

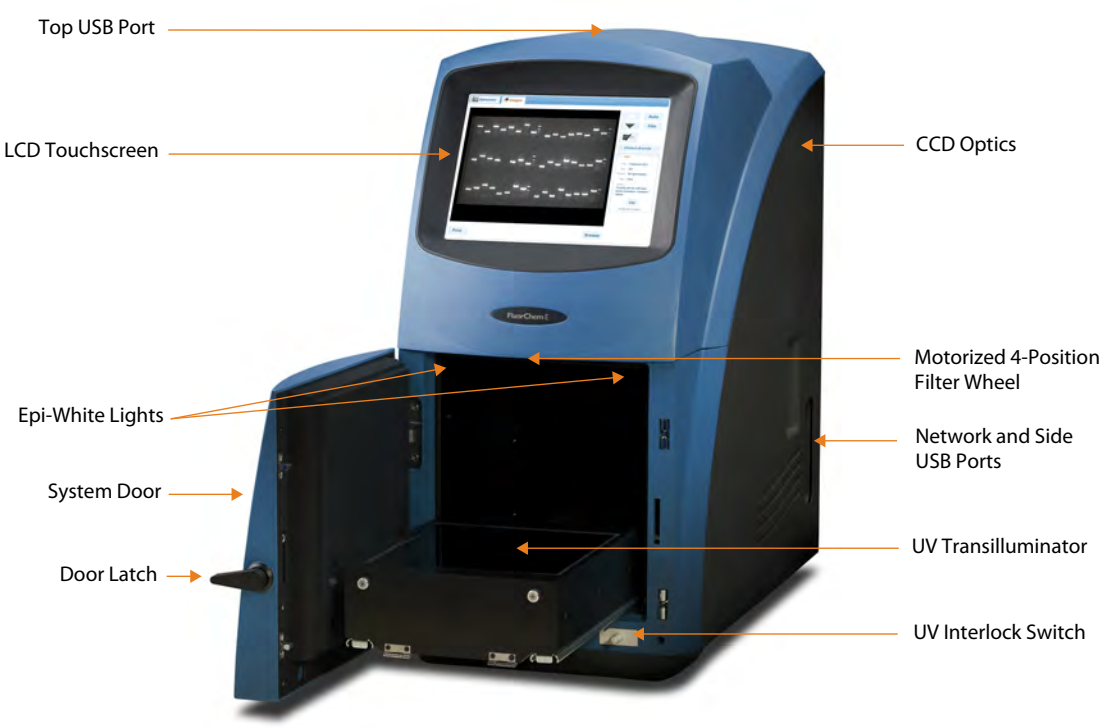
Chapter Overview

- Overview of FluorChem E System Components
- Compliance
- Safety Guidelines
- General Guidelines and Information
- System Specifications

Overview of FluorChem E System Components

FluorChem E is a high-performance imaging system for Western blot and gel imaging, and provides detection of chemiluminescent, colorimetric and UV-excited gels and blots. The individual hardware components of the FluorChem E system are described in this section.

FluorChem E Instrument



Integrated CCD Optics

The FluorChem E system uses a scientific grade 8.3 megapixel CCD detector cooled to -25 °C and a motorized lens for optimum resolution and acquisition speed. The optics are fully automated and require no manual user adjustments.

LCD Touchscreen

The 10.4" LCD touchscreen is used for direct control of the instrument through the FluorChem E User Interface. Image acquisition and review, system settings, and image file copy and printing options are accessed through commands on the touchscreen. Simply touch the screen to see menus or activate controls. ProteinSimple recommends using the stylus provided with the system to interact with the touchscreen.

System Door

The FluorChem E system door provides access to the inside of the instrument for loading of samples, gels and blots for imaging. To open the door, pull the door latch forward and down until it stops, then pull the door open. To close and lock the door, close the door and turn the door latch up and back to its stop position.

NOTE: The FluorChem E system door must be closed during image capture for proper instrument operation and user safety.

Epi-White Lights

Epi-white lights are also located at the top of the inside cabinet. The white light produced is used for sample positioning and imaging of colorimetric blots.

UV Transilluminator

An integrated UV transilluminator is located at the base of the inside cabinet and is used for excitation of stained DNA and protein gels. The unit slides out for easy sample loading. Power to the UV transilluminator is automatically disabled when the FluorChem E system door is open.

CAUTION

The FluorChem E system contains an integrated ultraviolet transilluminator. The system enclosure confines the radiation within the system and shields the user from exposure. Exposure to UV radiation can cause permanent damage to the eyes and skin.

The FluorChem E system UV transilluminator uses 302 nm bulbs. These bulbs can be replaced with 365 nm bulbs if needed. Please refer to "UV Transilluminator Bulb Replacement" on page 113 for information on replacing UV transilluminator bulbs.

UV Interlock Switch

A defeatable safety interlock switch located below the UV transilluminator automatically cuts off power to the transilluminator when the door is open. This eliminates any dangerous user exposure to UV light. As a safety precaution, the UV light will automatically turn off after three minutes when using the interlock switch.

When the system door is open, users can override the safety interlock switch and turn the UV light source on by inserting the supplied hardware key and turning clockwise. The switch can only be used when the **UV Light** button has been selected from the **Door Open Screen** in the user interface. For more information on how to use this option, please refer to "UV Light Override" on page 33.

CAUTION

Exposure to UV radiation can cause permanent damage to the eyes and skin. Always wear UV-protected eyewear and protective clothing over areas of skin that may be exposed to UV light when engaging the UV interlock switch override.

Filter Wheel

The FluorChem E system is equipped with a motorized 4-position filter wheel. The system comes with a pre-installed 590/50 nm orange filter commonly used for ethidium bromide or SYPRO orange applications. In addition, the following optional filters are also available:

- 620/40 nm (red) filter for SYPRO Red, SYPRO Ruby or Deep Purple
- 537/35 nm (green) filter for SYBR Green, SYBR Gold or SYBR Safe
- 460/40 nm (blue) filter for Hoeschst Blue, DAPI, ANS or Methylcoumarin

For information on how to insert optional filters into the filter wheel, please refer to "Installing Emission Filters" on page 116. If new filters are installed or existing filters are moved to different positions on the filter wheel, the filter settings in the **System Settings Screen** must be updated accordingly. To view or change filter settings, please refer to "Changing Filter Settings" on page 91.

Computer

The FluorChem E system contains an integrated computer with a 250 GB hard drive. Acquired images are automatically archived on the hard drive. Access to network and side USB ports is located on the right side panel, near the rear of the instrument.

Network Port

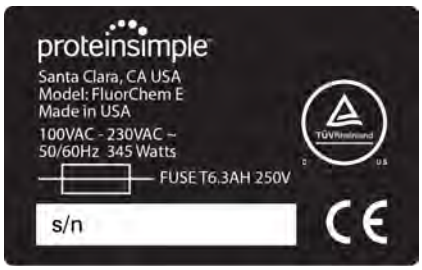
The 10/100/BASE-T Ethernet (RJ-45 connector) can be used to connect the FluorChem E system computer to a local network. Please see Chapter 9, *"Networking and Remote Access"* for information on accessing the system through a local network.

USB Ports

The FluorChem E system has two side and one top USB port. These ports can be used with USB flash drives for copying image files or to connect directly to a compatible USB thermal printer.

System Label

The FluorChem E system label is located on the rear panel of the instrument and includes the following information: ProteinSimple location, system model, power and fuse requirements, serial number and certification markings. An additional serial number label is located on the inside of the FluorChem E system door.



System Power

The power components consist of the main power switch, power entry and fuse. They are located on the system's rear panel.

!WARNING!

Only use the power supply cord provided with the instrument. If the power supply cord is damaged, please contact ProteinSimple Technical Support.

!WARNING!

Only use fuses rated for 250 V, 6 A.

!WARNING! SHOCK HAZARD

Disconnect power cord from the instrument power inlet to disconnect power to the instrument.

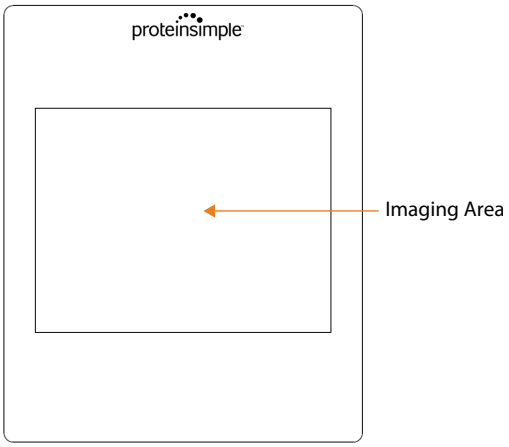
Sample Screens

The FluorChem E system is provided with three sample screens for imaging a wide range of applications. Sample screens are application specific and should be used as follows:

- **Black screen**: Chemiluminescent and epi white light imaging. Non-reflective and blocks residual UV light.

- **Clear screen**: UV imaging. Transmits UV light.
- **White screen**: Colorimetric gel imaging. Converts transilluminating UV light to white light for imaging of visible stains.
- **Blue screen (optional)**: Converts transilluminating UV light to blue light for fluorescent imaging of blue-excited stains.

An imaging area at the center of each screen assists users with sample positioning. Prior to imaging, samples should be placed on the appropriate sample screen and centered in the 16 cm x 12 cm imaging area. The screen is then placed directly on the UV transilluminator.



When not in use, sample screens should be stored in the screen holder provided with the FluorChem E system:

Compliance

The FluorChem E system complies with:

- **UL 61010-1:2004 R10.08**: Safety requirements for electrical equipment for measurement, control, and laboratory use - Part 1: General requirements (US)
- **EN 61010-1:2001**: Safety requirements for electrical equipment for measurement, control, and laboratory use - Part 1: General requirements (EU)
- **CAN/CSA 22.2 No. 61010-1-04**: Safety requirements for electrical equipment for measurement, control, and laboratory use - Part 1: General requirements (CA)
- **EN 61326-1:2006**: Electrical equipment for measurement, control and laboratory use. EMC Requirements. General requirements (EU)

Safety Guidelines

!WARNING!

If the instrument is not used as specified by the manufacturer the overall safety will be impaired.

!WARNING!

If the system is damaged and does not function properly, stop the instrument safely and contact ProteinSimple Technical Support immediately.

CAUTION

Avoid using the FluorChem E system in a manner not specified by ProteinSimple. While the system has been designed to protect the user, this protection may be impaired if the instrument is used improperly.

UV Transilluminator

CAUTION

The FluorChem E system contains an integrated ultraviolet transilluminator. The system enclosure confines the radiation within the system and shields the user from exposure. Exposure to UV radiation can cause permanent damage to the eyes and skin.

CAUTION

Under normal conditions, the temperature of the glass surface of UV transilluminator is safe to touch. However, if the system malfunctions, it is possible that the glass surface temperature could exceed 80 °C. Please exercise caution when touching the glass surface.

UV Interlock Switch

The FluorChem E system is equipped with a defeatable safety interlock switch which automatically cuts off the power to the transilluminator when the door is open. This eliminates any dangerous user exposure to UV light. As a safety precaution, the UV light will automatically turn off after three minutes when using the interlock switch.

General Guidelines and Information

System Operation: Notification of Intended Use

NOTE: The FluorChem E system is for research use only. Not for use in diagnostic or clinical procedures.

Lifting and Moving the System: Use Proper Lifting Precautions

IMPORTANT
Use caution when lifting or moving the FluorChem E system. The system weight is 75 lbs (34 kg). Two people are required to lift the system onto a laboratory bench. Proper lifting safety precautions must be taken.

System Specifications

Description	Specification
Dimensions	62 cm D x 62 cm H x 32 cm W (24.5 in x 24.5 in x 12.6 in)
Weight	34 kg (75 lbs)
Power Requirements	100 - 230 V AC, 50/60 Hz, self adjusting
Power Consumption	345 W
Operating Humidity Range	15 - 70% relative, non-condensing
Operating Temperature Range	15 - 25 °C (59 - 77 °F)
Detector	8 Megapixel scientific-grade CCD, 16-bit A/D, thermoelectrically cooled to -25 °C absolute and regulated
Optics	50-mm, f/1.4 motorized fixed lens
Field of View	16 cm x 12 cm (fixed)
Image output	16-bit PNG and 8-bit PNG

For indoor use only.

Chapter 4:

Getting Started

Chapter Overview

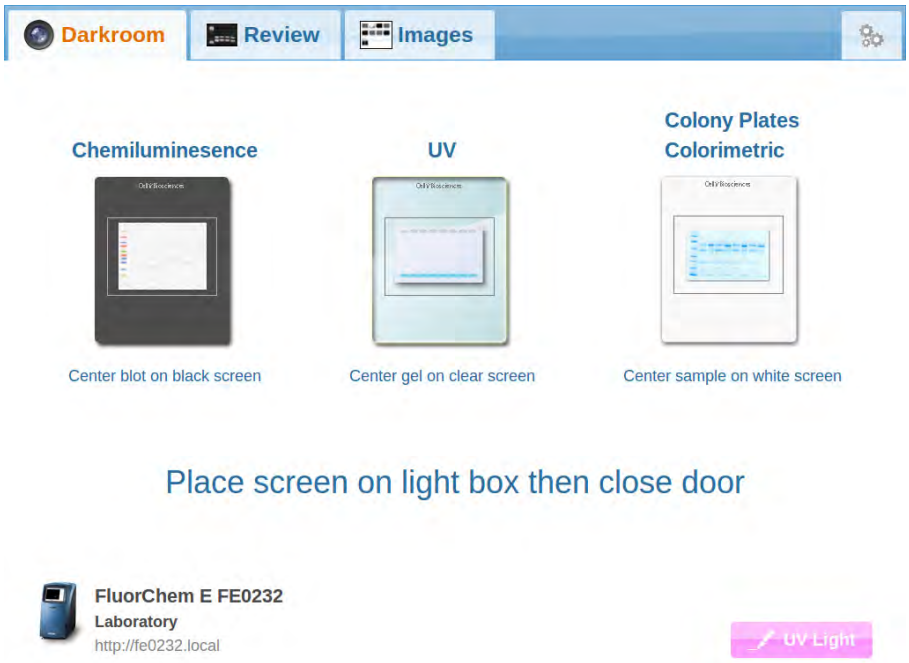
- Powering on the System
- Overview of FluorChem E User Interface
- Imaging Setup
- UV Light Override

Powering on the System

To power up the FluorChem E system, turn on the main power switch located at the rear of the instrument.

NOTE: When imaging chemiluminescent samples, allow the FluorChem E system to equilibrate for at least 30 minutes after power up. This ensures that the system optics have reached the optimum acquisition temperature prior to imaging. This equilibration time is not necessary if the system power remains on at all times.

When the system has finished booting up and is ready to use, the following screen will display:



NOTES:
ProteinSimple recommends leaving the FluorChem E system power on when not in use. Power the system down only when it will not be used for extended periods of time. Cycling the system power on a regular basis is not optimal for the system's integrated computer and cooled CCD optics.

The FluorChem E system must be powered on in order for a remote computer or device to connect and access the browser version of the FluorChem E User interface through a local network connection.

Overview of FluorChem E User Interface

This section will provide a general overview of the FluorChem E User Interface and touchscreen operation.

Using the Touchscreen

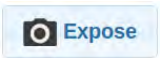
The FluorChem E User Interface is accessed through the touchscreen. Users can initiate acquisition, access protocols and images, and change system settings via touchscreen commands.

Tabs

Tabs are used to access particular screens in the FluorChem E User Interface. To view a screen, touch the screen tab.

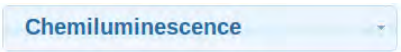
Buttons

Buttons are used for direct instrument control. To initiate a command, touch the button.

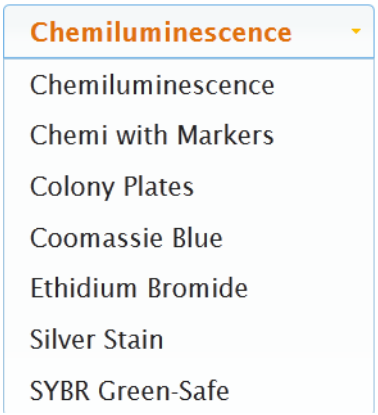


Menu Boxes

Menu boxes contain a list options that can be selected by the user.

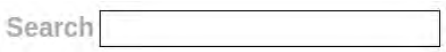


To view the items in the menu box drop down list, touch the box. To select an item in the menu, simply touch the item.

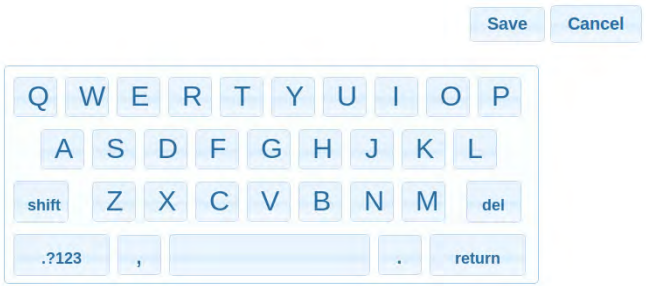


Text Boxes

Text boxes display information typically entered by the user.



To type an entry into a text box, touch the box. A keyboard or number pad will display on the screen and a cursor will be placed in the text box. Use the keyboard or number pad to type in the entry.



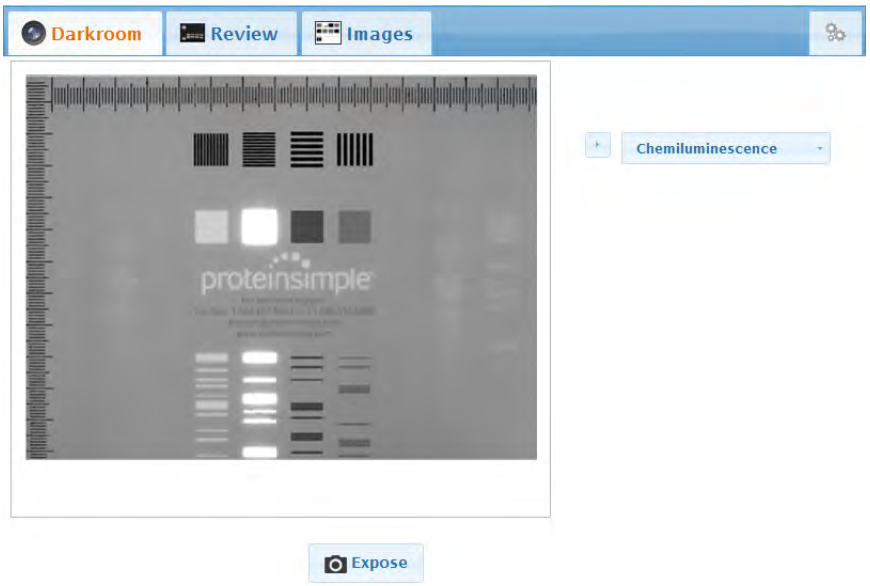
Use the **del** key to delete an existing entry. When text entry is complete, touch the **Save** button.

User Interface Screens

The FluorChem E User Interface has four main screens that are accessed through the following tabs: Darkroom, Review, Images and System Settings. Each screen can be accessed by selecting the tab on the touchscreen.

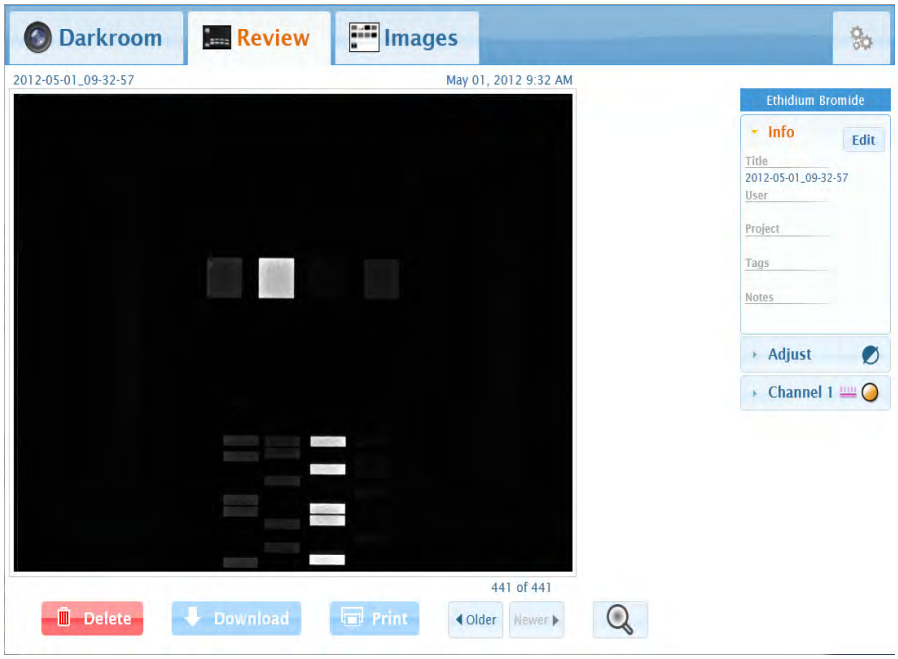
Darkroom Screen

Selecting the **Darkroom Tab** will display the **Darkroom Screen**. This screen is used for protocol setup and image acquisition.



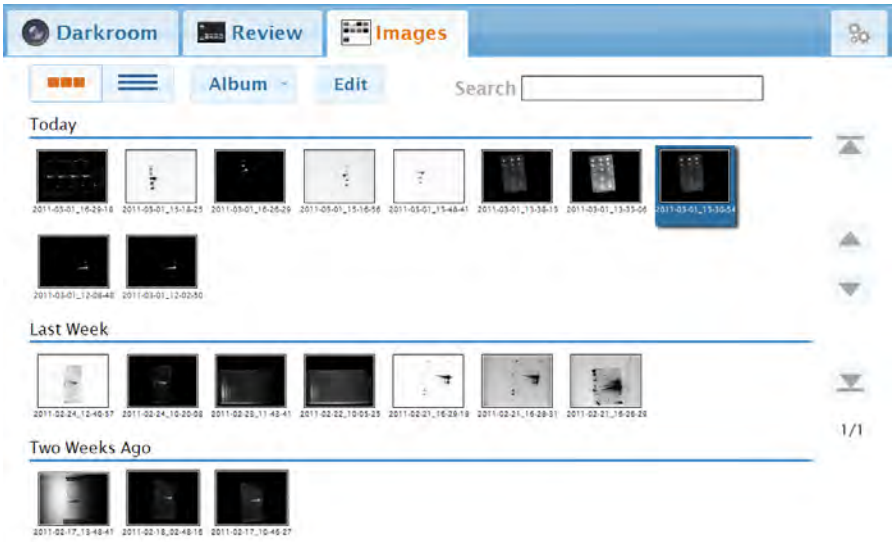
Review Screen

Selecting the **Review Tab** will display the **Review Screen**. This screen is used to view stored images and protocol information, enter or edit image details and make image enhancements. Images can also be deleted, copied and printed.



Images Screen

Selecting the **Images Tab** will display the **Images Screen**. This screen is used to browse saved images. Stored images can also be deleted and copied from this screen.



System Settings Screen

Selecting the tab on the far right will display the **System Settings Screen**. This screen displays system information and allows users to change the system name, date and time and focus settings as well as install software updates and run system tests. Dark masters, flat field calibration and system log files can also be generated.

Screen Saver

After a period of inactivity, the FluorChem E User Interface will automatically launch a screen saver to preserve the lifetime of the LCD touchscreen. When the screen saver is running the screen will be blank. Simply touch the screen to exit the screen saver mode and view the user interface.

Imaging Setup

Prior to image acquisition, samples must be placed on the appropriate sample screen and positioned properly on the UV transilluminator.

Sample Screens

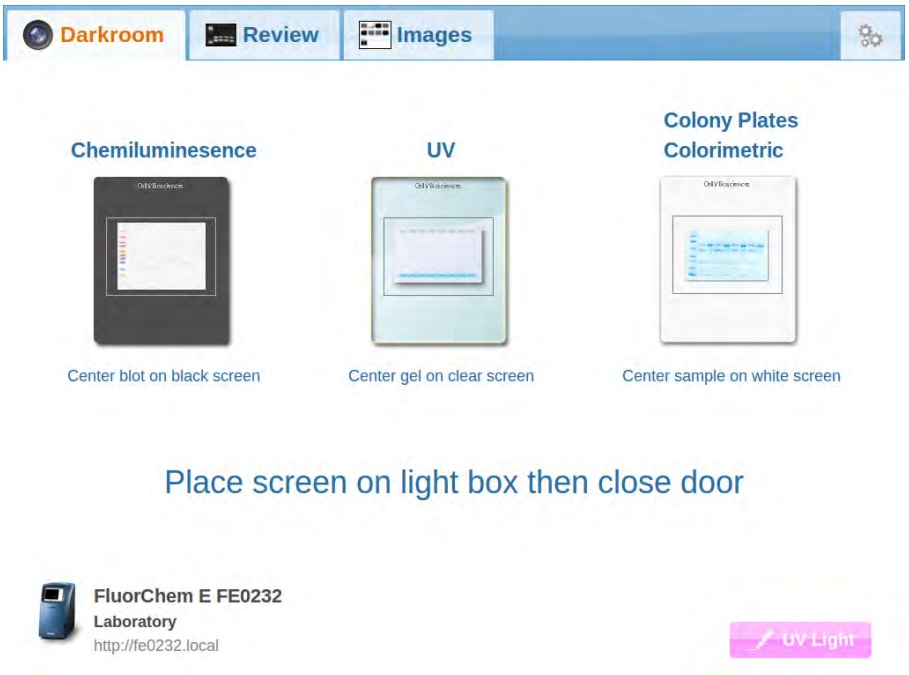
The FluorChem E system is provided with the following screens for sample imaging. The screen used depends on the application:

- **Black screen**: Chemiluminescent and epi white light imaging
- **Clear screen**: UV-excited gel imaging
- **White screen**: Colorimetric gel imaging and colony plates
- **Blue screen (optional)**: Blue-excited fluorescent gel imaging

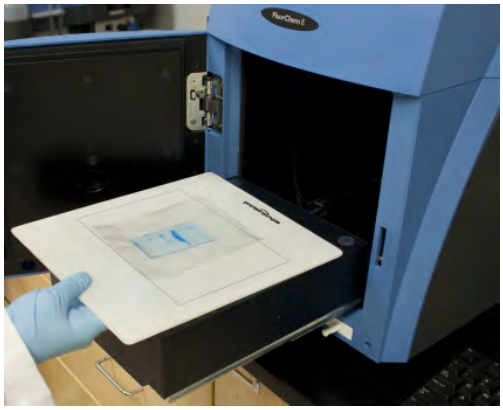
Sample Positioning

To position samples in the FluorChem E system:

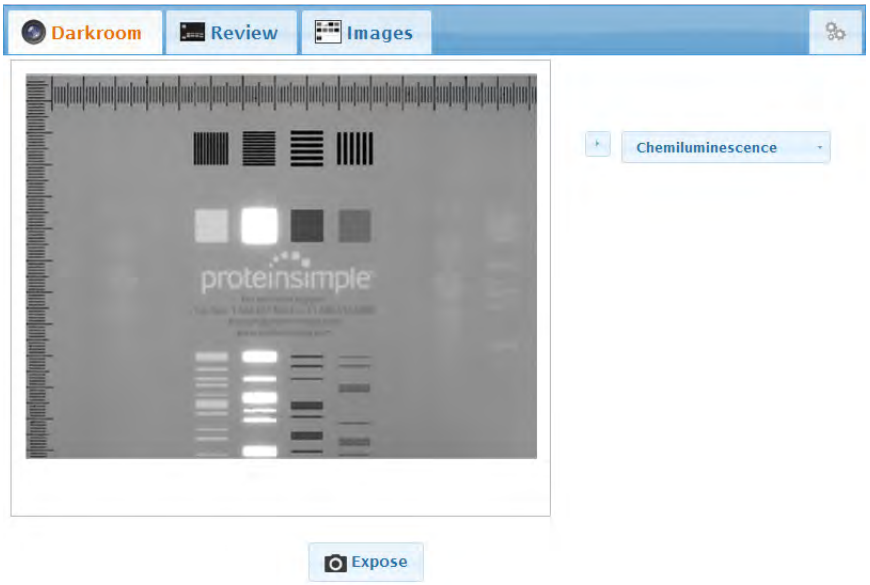
1. Place the sample on the appropriate sample screen and center it on the imaging area.
2. Open the system door. The **Door Open Screen** will display:



3. Place the screen on the UV transilluminator.



4. Close the system door. The FluorChem E system will automatically take an epi-white light image of the sample.



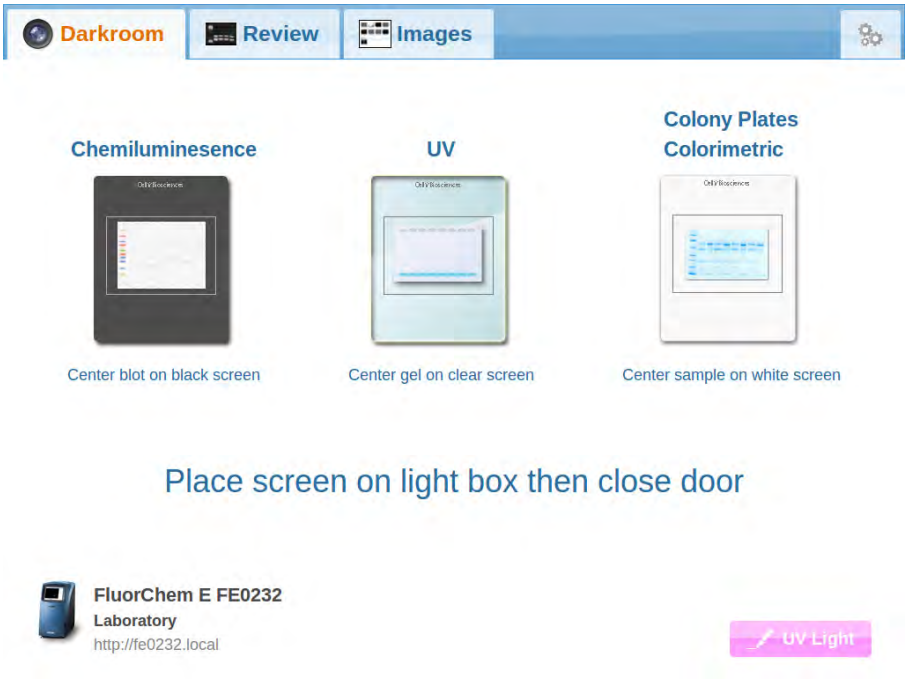
5. Ensure that the area of interest in the blot or gel has been captured in the image. If not, open the system door and reposition the sample in the imaging area of the sample screen as needed. The FluorChem E system will take a new white light image each time the system door is closed.

The sample is now ready to be imaged.

UV Light Override

If necessary, users can override the safety interlock switch and turn on the UV transilluminator manually. To do this:

1. Open the system door. The **Door Open Screen** will display:



2. Select the **UV Light** button on the touchscreen. The following UV light dialog box will display:

3. Insert key and turn clockwise.

NOTE: As a safety precaution, the UV light will automatically turn off after three minutes when using the interlock switch.

CAUTION

Exposure to UV radiation can cause permanent damage to the eyes and skin. Always wear UV-protected eyewear and protective clothing over areas of skin that may be exposed to UV light when engaging the UV interlock switch override.

4. When finished, turn off the UV transilluminator by turning the key couterclockwise and removing. Then select the **Off** button to exit the UV light dialog box.

Chapter 5:

Image Acquisition

Chapter Overview

- Overview of the Darkroom Screen
- Image Acquisition
- Sample Imaging

Overview of the Darkroom Screen

Selecting the **Darkroom Tab** will display the **Darkroom Screen**. This screen is used to view protocols and channel settings and for image acquisition.

Imaging Setup

The **Setup** page will be displayed whenever the **Darkroom Tab** is selected. Users can select protocols, change channel settings and start image acquisition in the **Setup** page.

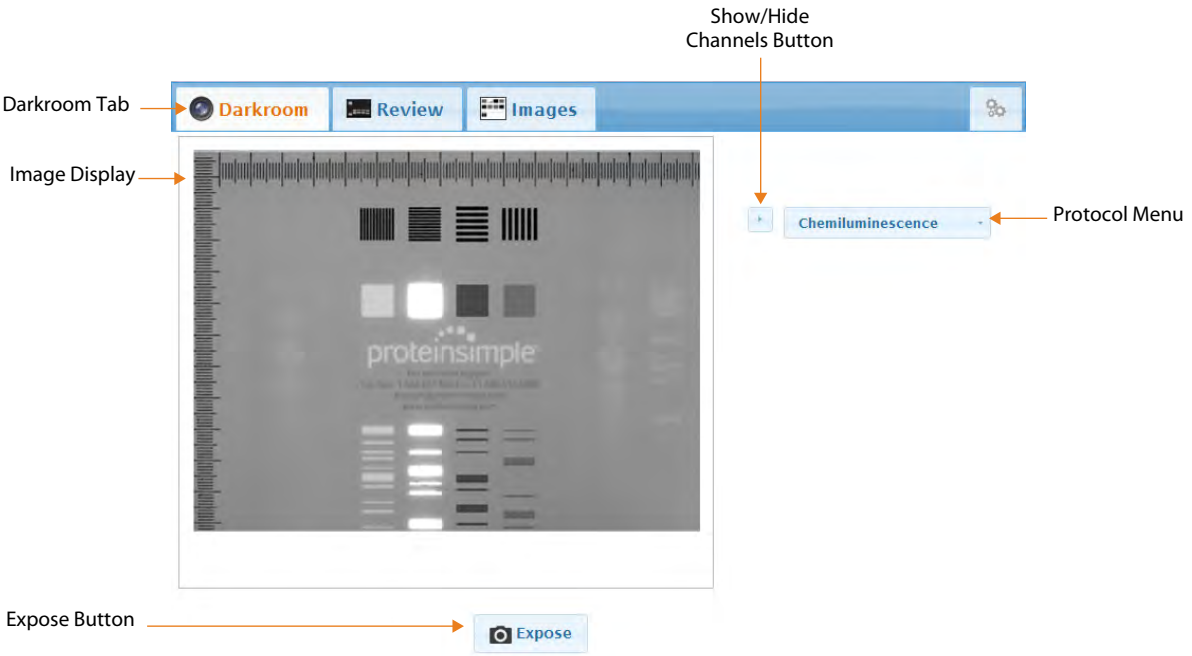
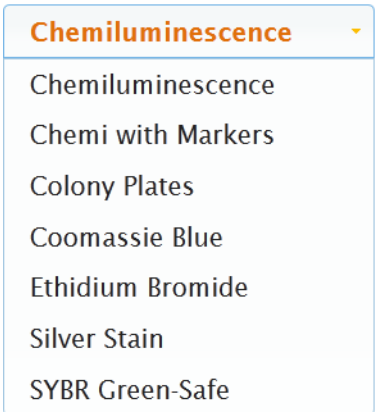


Image Display

The **Image Display** area will display a white light image that is automatically taken to help with sample positioning prior to image acquisition.

Protocol Menu

This menu allows users to select from a set of pre-programmed protocols. Protocol options can be viewed by selecting the **Protocol Menu** box.

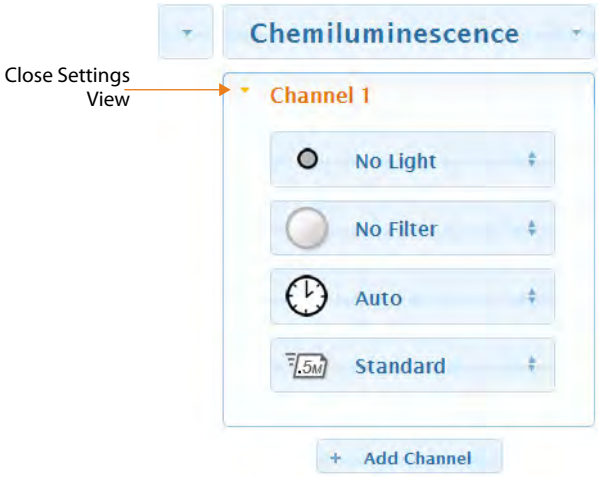


Show/Hide Channels

Selecting the **Channel Show/Hide** button to the left of the **Protocol Menu** box will display the channels used with the current protocol and/or added by the user. Users have the option of adding channels by selecting the **Add Channel** button. Selecting the **Channel Show/Hide** button again will hide the channel display. For more information on channels, please see "Multi-Channel Imaging" on page 45.

Channel Settings Menu

The **Channel Settings Menu** box allows users to view and change light, filter, exposure and resolution settings for each channel. These settings can be viewed by selecting the **Channel Settings Menu** box.



The **Channel Settings Menu** box displays icons representing the light and filter settings used for each channel. These icons will automatically update whenever channel settings are changed.

To close the settings view, select the orange arrow or channel number in the upper left of the **Channel Settings Menu** box.

Expose Button

The **Expose** button starts image acquisition.

Selecting a Protocol

Protocol selection depends on the sample being imaged. Table 5-1shows a list of pre-programmed protocols along with the light setting, filter setting and the recommended sample screen for each.

Protocol	Channel	Light	Filter	Sample Screen
Chemiluminescence	CH 1	No light	No filter	Black
Chemi with Markers	CH 1 CH 2	No light Epi White	No filter Orange 590 nm	Black
Colony Plates	CH 1	Trans UV	Orange 590 nm	White
Coomassie Blue	CH 1	Trans UV	Orange 590 nm	White
Ethidium Bromide	CH 1	Trans UV	Orange 590 nm	Clear
Silver Stain	CH 1	Trans UV	Orange 590 nm	White
SYBR Green/Safe	CH 1	Trans UV	Green 537 nm	Clear
SYPRO Ruby	CH 1	Trans UV	Red 620 nm	Clear

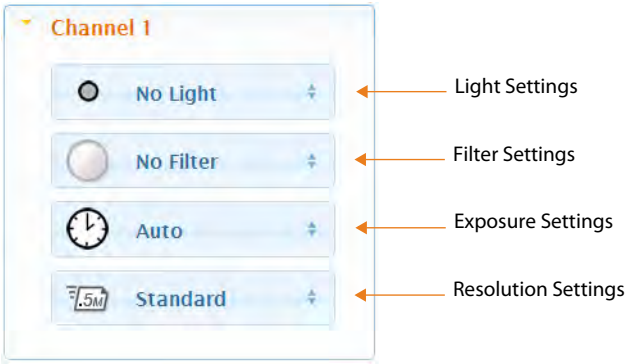
Table 5-1: *FluorChem E system pre-programmed protocols.*

The protocols available in the **Protocol Menu** depend on the filters installed in the FluorChem E system. Only protocols for the installed filters will be available for selection in the drop down menu.

Changing Channel Settings

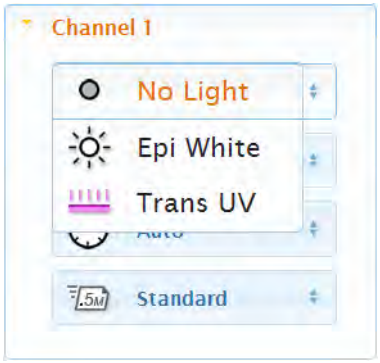
Light, filter, exposure and resolution settings for each channel used in a pre-programmed protocol can be viewed and changed by selecting the **Channel Settings Menu** box. These settings can be changed prior to acquiring an image.

NOTE: Changes to default channel settings are not permanently saved. When settings are modified, the new selections are used for image acquisition until the FluorChem E system door is opened. At that time, all channel settings will return to the default selections specified by the protocol.



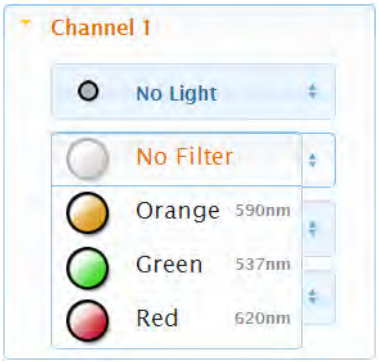
Light Settings

To change the light source, select the **Light Settings** box and select the desired option:



Filter Settings

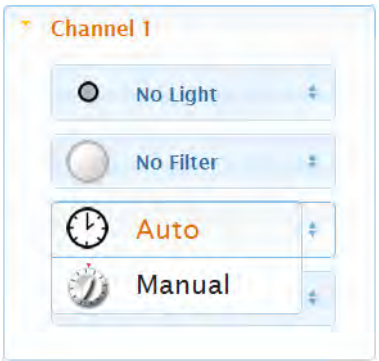
To change the filter, select **Filter Settings** box and select the desired option:



*NOTE: Up to three filters can be installed in the FluorChem E system. The **Filter Settings** box will only display installed filters. For information on how to add or change the filters listed in the **Filter Settings** box, please refer to "Changing Filter Settings" on page 91.*

Exposure Settings

To change exposure settings, select the **Exposure Settings** box and select one of the following options:



Auto Exposure

When **Auto** is selected in the protocol, the FluorChem E system will take successively longer exposures during image acquisition until the optimal exposure is reached. However, users can select the **Stop** button at any time to save and display the last completed image exposure taken.

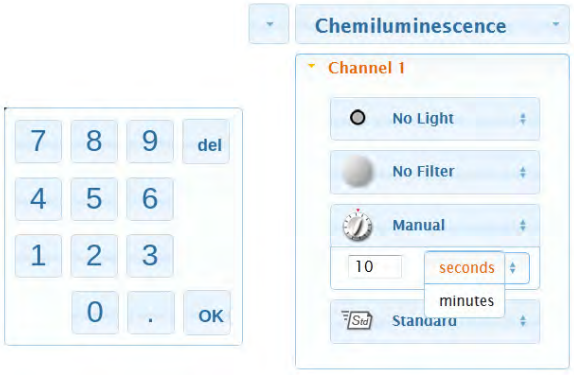
Manual Exposure

When **Manual** is selected in the protocol, the FluorChem E system will take one image at the exposure time entered. To enter an exposure time, select the time box. A numeric keypad will appear to allow the user to enter a time setting.

Any value between 50 ms and 3 hours can be used. Settings are displayed in either seconds or minutes. To enter a setting of less than 1 second, select the decimal point first.

Example 1: Manual Exposure Time of 50 ms

Enter .05 for a manual exposure time of 50 ms. The time displayed will be 0.05 seconds:

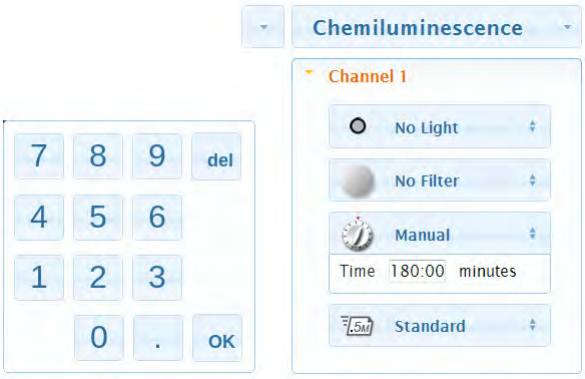


Example 2: Manual Exposure Time of 90 seconds

Either 130 or 90.00 can be entered for a manual exposure time of 90 seconds. The time displayed will either be 1:30 minutes or 90 seconds depending on how the time was entered:

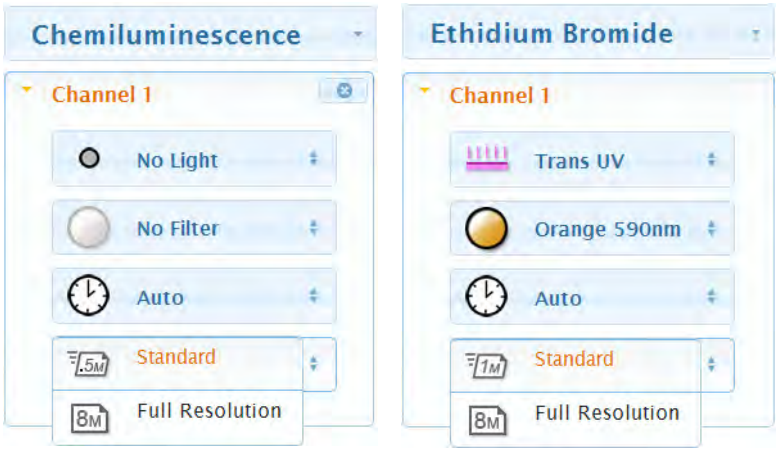
Example 3: Manual Exposure Time of 3 hours

Enter 18000 for a manual exposure time of 3 hours. The time displayed will be 180 minutes:



Resolution Settings

To change resolution, select the **Resolution Settings** box and select one of the following options:



Standard Resolution

When **Standard Resolution** is selected in the protocol, the FluorChem E system will acquire the image with a 4 x 4 pixel bin (832 x 626 pixels) for chemiluminescence protocols and a 3 x 3 pixel bin (1109 x 835 pixels) for all other protocols. **Standard Resolution** is the default and recommended setting for all imaging protocols. For chemiluminescence imaging, this setting significantly decreases the exposure time required to visualize samples with low signal intensities.

Full Resolution

When **Full Resolution** is selected in the protocol, the FluorChem E system will acquire the image using full 8 megapixel resolution (3326 x 2504 pixels). **Full Resolution** is only recommended when publication quality and large printouts (e.g. poster size) are required, or to resolve extremely small feature sizes (<150 microns). Selecting **Full Resolution** significantly increases the exposure time required compared to **Standard Resolution**.

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*NOTE: Selecting **Full Resolution** will result in increased exposure time. This increased exposure time can be very significant, particularly when imaging low abundant chemiluminescent targets.*

Multi-Channel Imaging

FluorChem E provides optional multi-channel image capture on up to four channels per acquisition when needed for specific applications. Each channel can then be subsequently overlaid or stacked in the **Review Screen** for visualization of images such as:

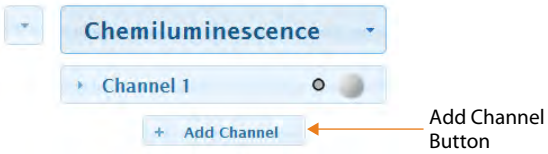
- Colorimetric markers on chemiluminescent blots
- Chemiluminescent blots at various exposure times

Adding Channels

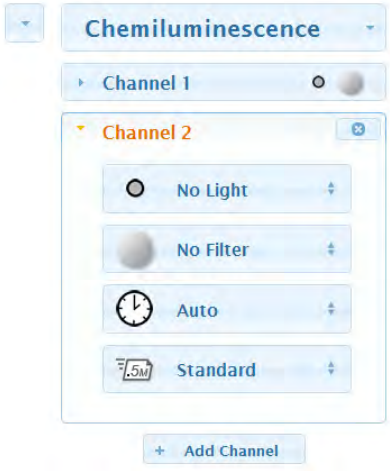
Once a protocol is selected, a total of up to four image capture channels can be added prior to acquisition. A common application requiring additional channels would be when multiple manual chemiluminescent exposures are desired.

All protocols must contain at least one channel. The **Channel Settings Menu** box will display icons representing light and filter settings used for each channel. These icons will automatically update whenever channel settings are changed.

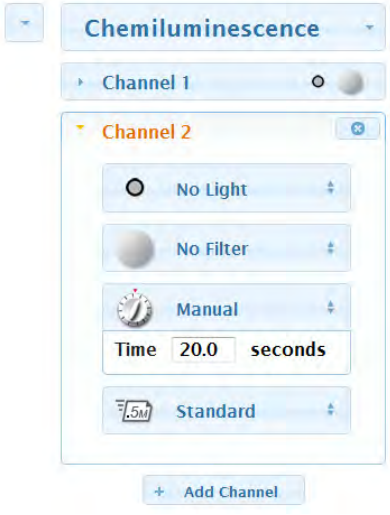
To add a channel, select the **Add Channel** button.



The new channel added will by default display the same settings as the previous channel.



Change light, filter, exposure and resolution settings as desired.

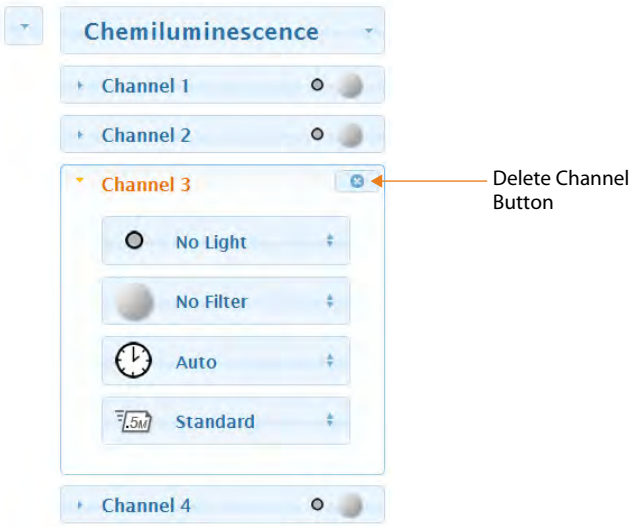


Continue adding channels and changing settings as needed. A total of up to four channels can be added. Once a fourth channel is added, the **Add Channel** button will no longer display.

*NOTE: Channels that were added manually will be deleted automatically if a new protocol is selected in the **Protocol Menu** box.*

Deleting Channels

To delete a channel, select the **Channel Settings Menu** box, then select the **Delete Channel** button in the upper right corner.



After a channel has been deleted, the remaining channels will automatically renumber accordingly.

NOTE: Imaging protocols must contain at least one channel. Channel 1 in a single-channel protocol will not be available for deletion.

Image Acquisition

The **Acquire** page will display in the **Darkroom Screen** once image acquisition has started. Users can monitor and stop image acquisition, view protocol and channel settings and channel image data in the **Acquire** page.

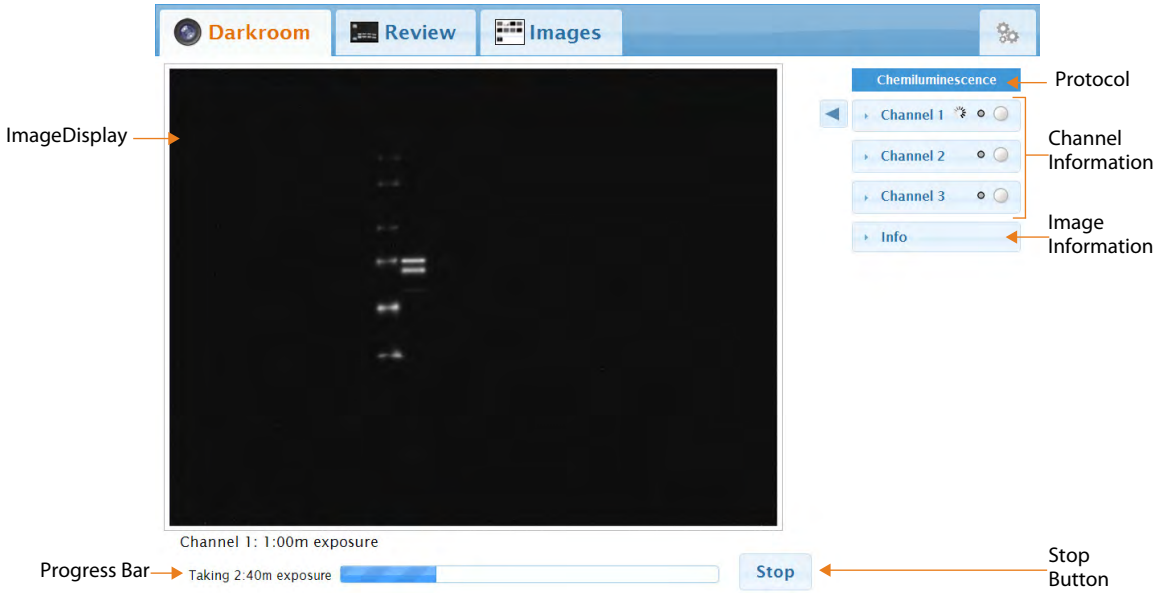


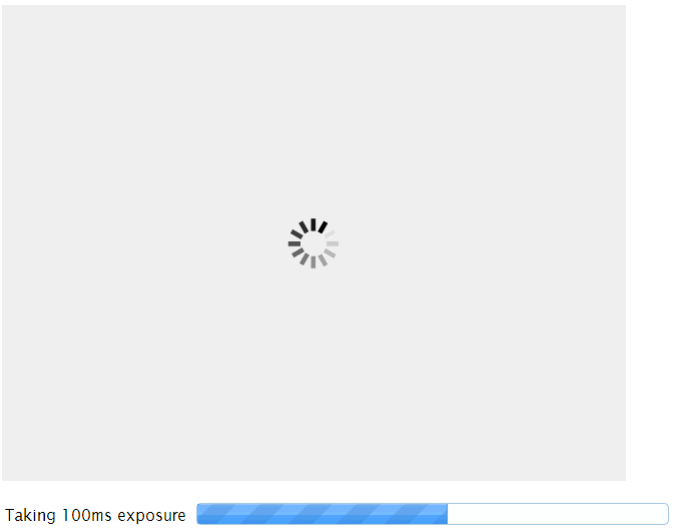
Image Display

The **Image Display** area will display the image as it is acquired. The channel number and exposure time will be displayed under the **Image Display**.

When a channel uses **Auto** expose, the **Image Display** and exposure time will refresh each time a new exposure is taken during image acquisition.

Channel 2: 150ms exposure

When a channel uses **Manual** expose, the **Image Display** and exposure time will not present data until the image has been taken at set exposure time. The acquisition indicator will display while imaging is in progress.



Progress Bar

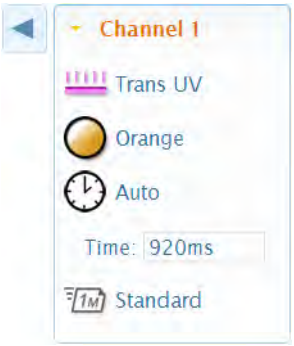
The **Progress Bar** will indicate the FluorChem E system status during image acquisition and will refresh as each exposure is taken.

Protocol Information

The **Protocol** box will display the protocol currently being used for image acquisition.

Channel Information

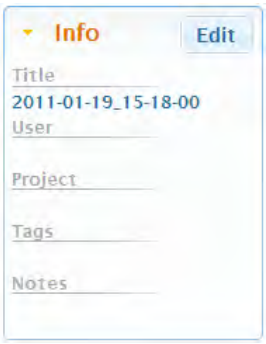
The **Channel Settings** box(es) will display the individual channel settings used for image acquisition. To view light, filter, exposure and resolution settings select the **Channel Settings** box:



When channel acquisition is in progress, an acquisition indicator will display in the **Channel Settings** box. The **Image Show/Hide** button to the left of each **Channel Settings** box indicates the channel image data currently displayed. In the example below, Channel 1 image data is displayed and Channel 2 acquisition is in progress. Please see "Changing the Channel Image Display" on page 51 for more information on viewing channel image data.

Image Information

Selecting the **Info** box will display the initial image information. By default, the **Title** entry will be the date and time that the image was acquired. During or after acquisition is complete, the **Title** can be edited and the user, project, tags and notes for the image file can also be entered:



Stop and Stop All Buttons

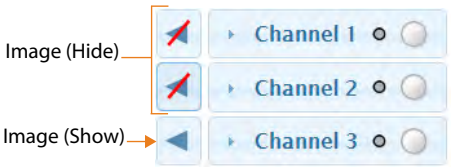
The **Stop** button halts image acquisition only for the channel that is currently being acquired. It will not halt acquisition for any subsequent channels. If the protocol contains more than one channel, the following progress bar and **Stop All** button will display:

Select the **Stop All** button to halt acquisition for all channels. Otherwise, acquisition will continue for the remaining channels.

When a protocol uses **Auto** expose, selecting the **Stop** button will halt image acquisition and save the last completed exposure as the final sample image. When a protocol uses **Manual** expose, selecting the **Stop** button will halt image acquisition and discard any exposure data.

Changing the Channel Image Display

During acquisition, the **Image Display** will update to show image data for each channel as it is acquired. However, image data for any channel can be displayed by selecting the **Image Show/Hide** button next to the **Channel Settings** box. The button can be toggled between **Show** (arrow only) and **Hide** (red line through arrow).



NOTE: Image data for only one channel at a time can be displayed during the acquisition process.

To view image data for a different channel, toggle the **Image Show/Hide** button next to the desired **Channel Settings box** to **Show**. The image data for the selected channel will be shown, and all other channel image data will be hidden. The **Image Show/Hide** button(s) for the remaining channels will change to **Hide** automatically.

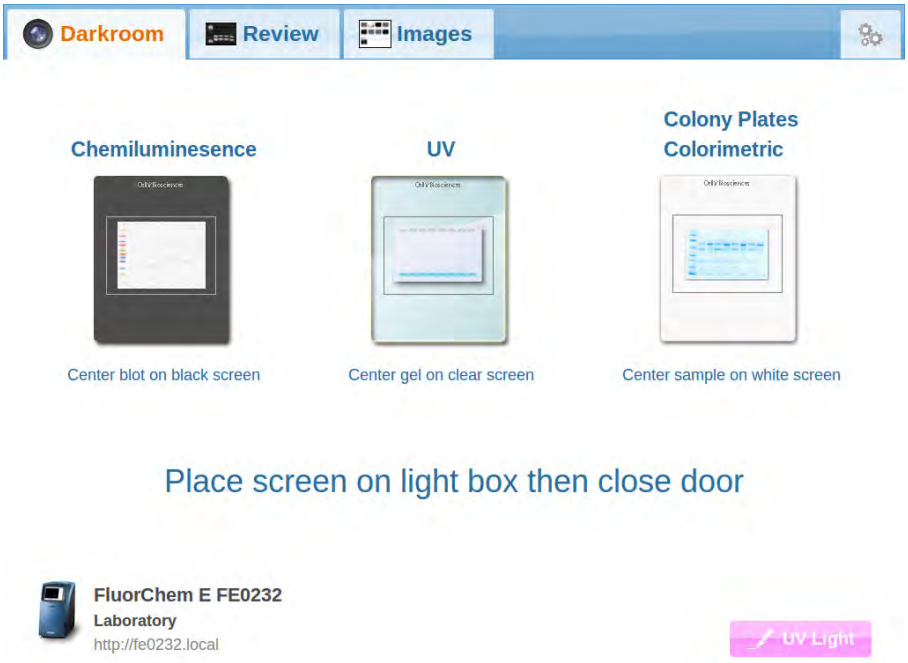
Sample Imaging

NOTES:
When imaging chemiluminescent samples, allow the FluorChem E system to equilibrate for at least 30 minutes after power up. This ensures that the system optics have reached the optimum acquisition temperature prior to imaging. This equilibration time is not necessary if the system power remains on at all times.

If the system optics temperature is not at optimum (-25 °C) when acquisition is started, a message will be displayed providing users with the option to either cancel or proceed with the acquisition.

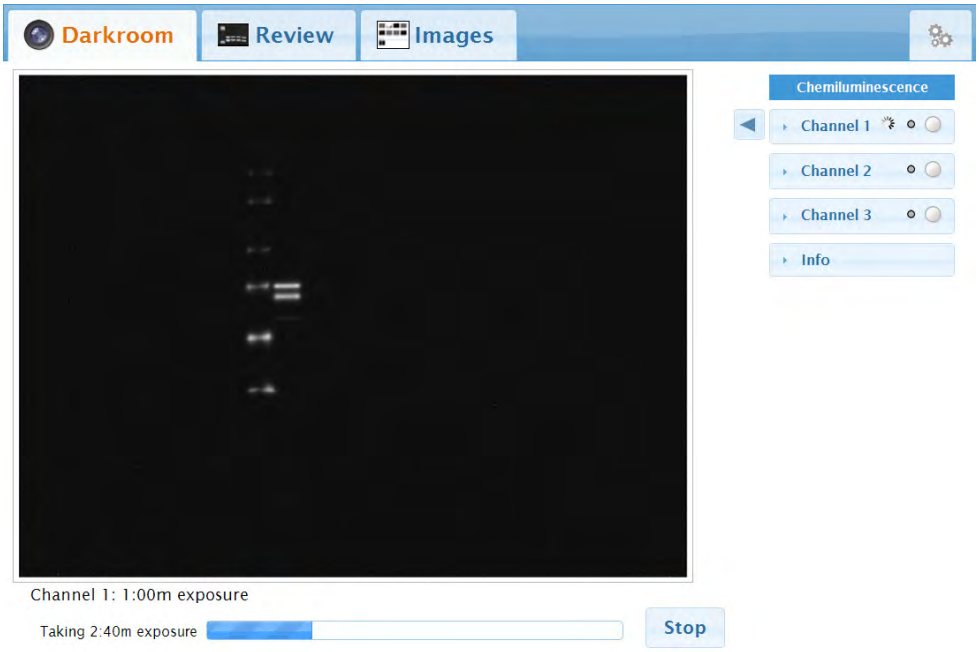
To image samples using the FluorChem E system:

1. Place the sample on the appropriate sample screen and center it on the imaging area.
2. Open the system door. The **Door Open Screen** will display:



3. Place the screen on the UV transilluminator.
4. Close the system door. The FluorChem E system will take an epi-white light image of the sample automatically.
5. Ensure that the area of interest in the blot or gel has been captured in the image. If not, open the system door and reposition the sample in the imaging area of the sample screen as needed. The FluorChem E system will take a new white light image each time the system door is closed.
6. Select a protocol from the **Protocol Menu,** change settings in the **Channel Settings Menu**, make changes to image information in the **Info** box, and add channels if desired.

Select the **Expose** button to start image acquisition. The **Acquire** page will display and Channel 1 acquisition will begin:



For channels using **Auto** expose, image acquisition will continue automatically until the correct exposure is determined. If the channel is using **Manual** expose, image acquisition will halt when the exposure time entered in the channel settings is reached. Image acquisition can be halted at any time by selecting the **Stop** button.

NOTES:
*Multi-channel acquisition will occur sequentially. Selecting the **Stop** button during a multi-channel acquisition halts acquisition only for the channel being acquired. It will not halt acquisition for any subsequent channels. To halt acquisition on all channels, select the **Stop All** button that becomes available after **Stop** is selected. Image data will only be available for those channels that completed acquisition.*

When the Chemi with Markers protocol is used for image acquisition, the chemiluminescent image of the sample (Channel 1) will display in grayscale and the white light image of the markers (Channel 2) is displayed in red to optimize visualization.

When acquisition is complete, all channel data is processed and final image(s) will be displayed in the **Review Tab** which will be described in detail in the next chapter.

Chapter 6:
Image Review

Chapter Overview

- Overview of the Review Screen
- Entering Image Information
- Channel Image Display and Overlay
- Making Image Adjustments
- Image Zoom
- Viewing Pixel Values
- Browsing Images
- FluorChem E System File Information
- Copying, Printing and Deleting Images

Overview of the Review Screen

Selecting the **Review Tab** will display the **Review Screen**. This screen is used to view images, protocol and channel settings, enter or edit image information and make any needed enhancements. Stored images can also be deleted, copied and printed. The **Review Screen** will be displayed automatically when image acquisition has completed.

NOTES:
Images are displayed overlaid by default for protocols that acquired images on multiple channels.

For images acquired using the Chemi with Markers protocol, the white light image of the markers (Channel 2) is displayed in red to optimize visualization. Images for channels acquired using all other protocols and/or light sources will display in grayscale.

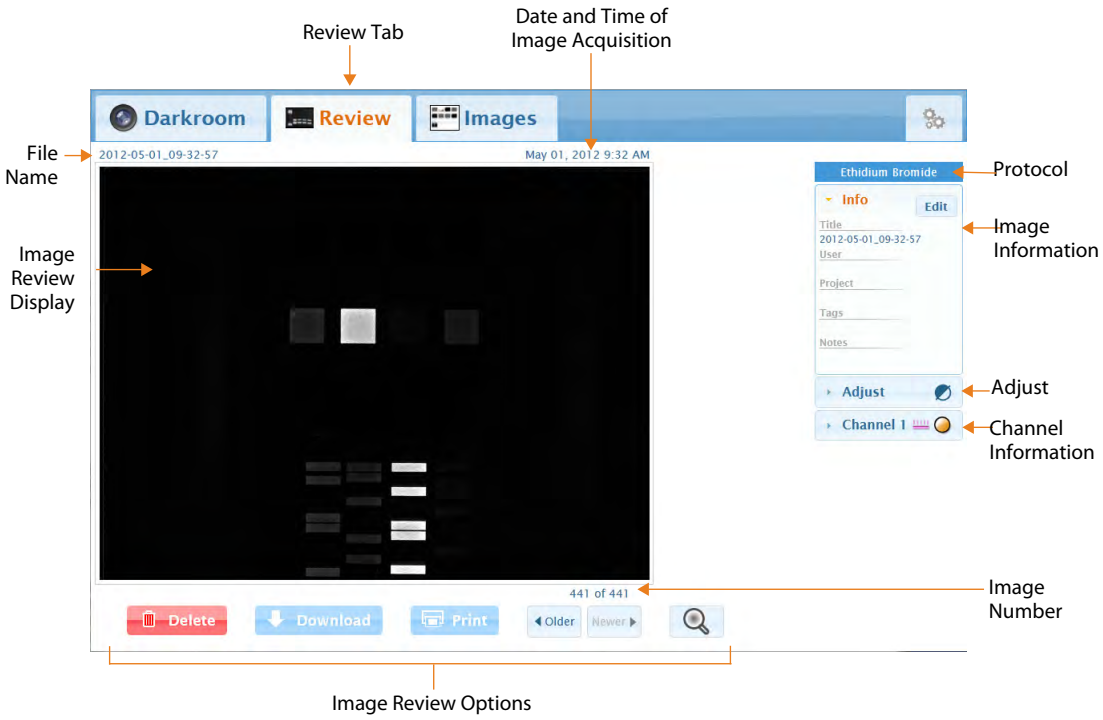


Image Review Display

The **Image Review Display** will display saved sample images. The file name is located above the display on the left, and the date and time of acquisition is located above the display on the right. The image number is shown below the display on the right.

Image Enhancement Tools

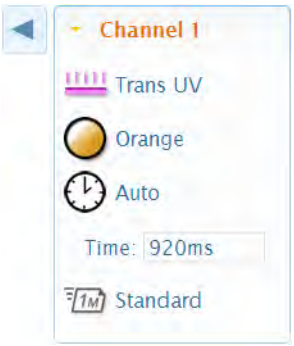
The enhancement tools on the **Review** page under **Adjust** include **Brightness Controls**, **Exposures View** and **Invert**. The use of these tools will be discussed in more detail later in this chapter.

Protocol Information

A copy of the acquisition protocol is stored with all images. The **Protocol** box will display the name of the acquisition protocol used for the image currently displayed.

Channel Information

The **Channel Settings** box(es) will display the individual channel settings used for image acquisition. To view light, filter, exposure and resolution settings select the **Channel Settings** box:

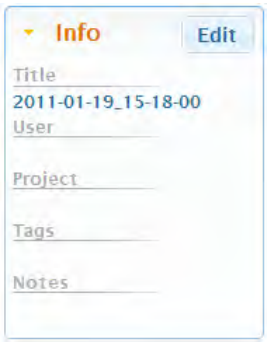


*NOTE: If the **Auto Expose** setting was used and image acquisition was stopped by the user, **Auto Stopped** will display as the exposure setting in the **Channel Settings** box.*

The **Image Show/Hide button** to the left of each **Channel Settings** box indicates what channel image data is currently displayed. In the example shown next, image data for Channels 1 and 3 are shown, and Channel 2 image data is hidden. Please see "Channel Image Display and Overlay" on page 60 for more information on viewing and overlaying channel image data.

Image Information

Any sample and image information entered by the user is also stored with all images. This information can be viewed by selecting the **Info** box:



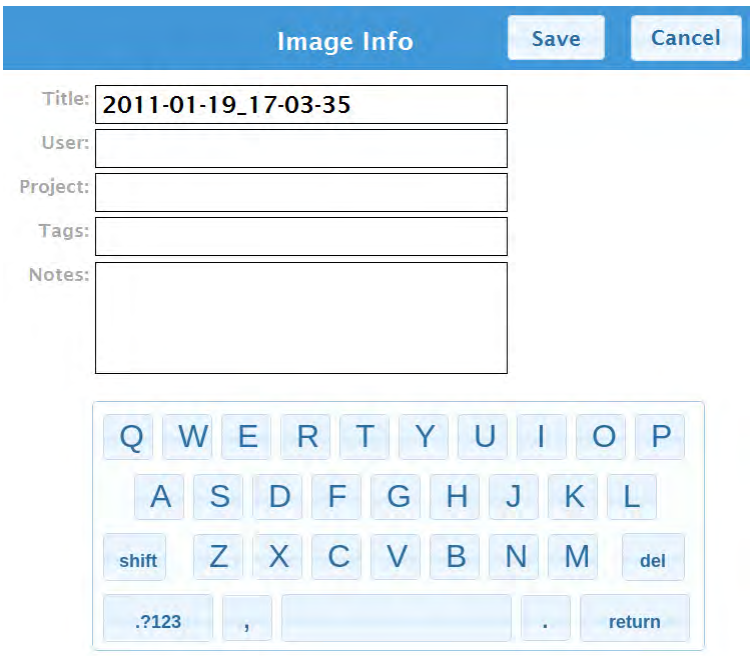
*NOTE: If the **Auto Expose** setting was used and image acquisition was stopped by the user, **Auto Stopped - see protocol details** will display as the title in the **Info** box.*

Image Review Options

The buttons located under the **Image Display** allows users to browse between files currently displayed on the **Images Tab** as well as **Delete**, **Copy** and **Print** images. In addition, the **Zoom** feature provides image magnification.

Entering Image Information

To enter sample image information, select the **Info** box. Then select the **Edit** button. The **Image Info** page will display:



The image title default is the date and time of acquisition and can be edited by the user. Enter text in the title, user, project, tags and notes fields as desired using the touchscreen keyboard.

When text entry is complete, select **Save**. Information entered will be saved with the final image and will also be searchable in the **Browse** page of the **Images Screen**. Select **Cancel** to exit the **Image Info** page.

Channel Image Display and Overlay

In the **Review Screen**, image data for all channels acquired can be viewed individually or overlaid. Channel overlay is ideal for visualization of colorimetric markers on chemiluminescent blots and chemiluminescent blots at various exposure times (stacking).

Channel image data can be displayed or hidden by selecting the **Image Show/Hide** button next to the **Channel Settings** box. The button can be toggled between **Show** (arrow only) and **Hide** (red line through arrow).

---

*NOTE: When multiple channels are acquired, channel image data will be displayed overlaid by default when the **Review Screen** is presented at the completion of acquisition.*

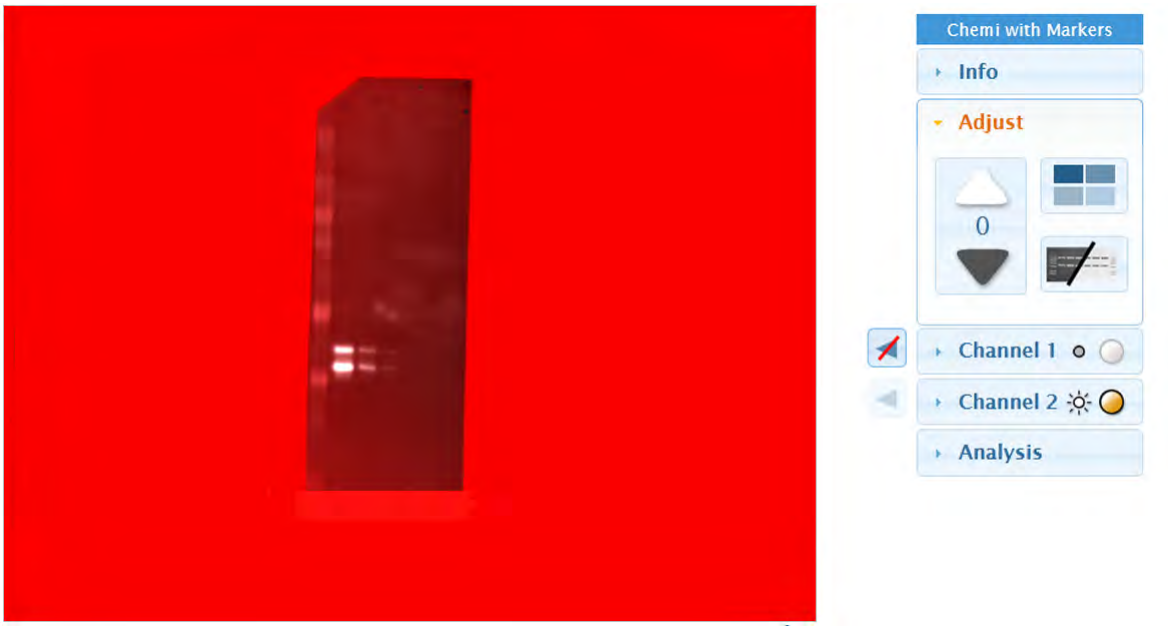
Viewing Image Data for One Channel

To view image data for a single channel, toggle the **Image Show/Hide** button next to the desired **Channel Settings** box to **Show**. Next, toggle the **Image Show/Hide** button(s) for the remaining channels to **Hide**. In the example that follows, image data for only Channel 1 is shown, Channel 2 image data is hidden.

Overlaying Channel Image Data

To overlay image data for multiple channels, toggle the **Image Show/Hide** buttons next to the desired **Channel Settings** boxes to **Show**. If needed, toggle the **Image Show/Hide** button(s) to **Hide** for any unwanted channels. When multiple channels are toggled to **Show**, the channel image data will be overlaid automatically. In the next example, image data for Channel 1 and Channel 2 are shown overlaid.

NOTE: When the Chemi with Markers protocol is used for image acquisition, the chemiluminescent image of the sample (Channel 1) will display in grayscale and the white light image of the markers (Channel 2) is displayed in red to optimize visualization.



Making Image Adjustments

NOTES:
Image adjustments are automatically saved to the image file as the enhancement settings are changed.

Image adjustments only change the way the image is visualized. They do not alter the original image data or affect quantitative results.

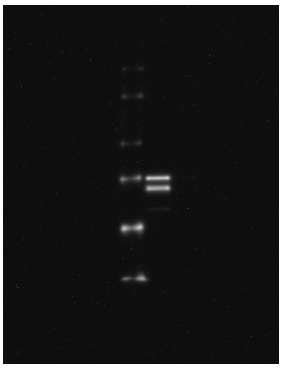
Brightness Controls

The **Brightness Controls** allow users to make images lighter or darker via a non-linear gamma adjustment. To adjust image brightness, select the up or down arrows. The setting control indicator will increase or decrease as adjustments are made. All images have a default setting of zero.

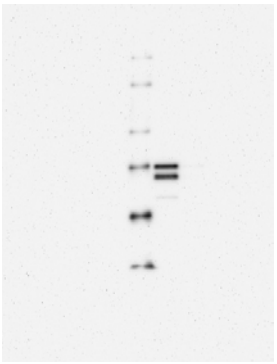
Invert

The **Invert** function inverts the gray levels of the displayed image, converting a positive image to negative or vice versa. For example, an image with black bands on a white background will be converted to an image with white bands on a black background. Selecting the button a second time returns the image to its original form.

NOTE: Image adjustments only change the way the image is visualized. They do not alter the original image data or affect quantitative results. Bands in the image will have the same quantitative densities in both the original and inverted versions of the image.



Chemiluminescent Image
Before Invert

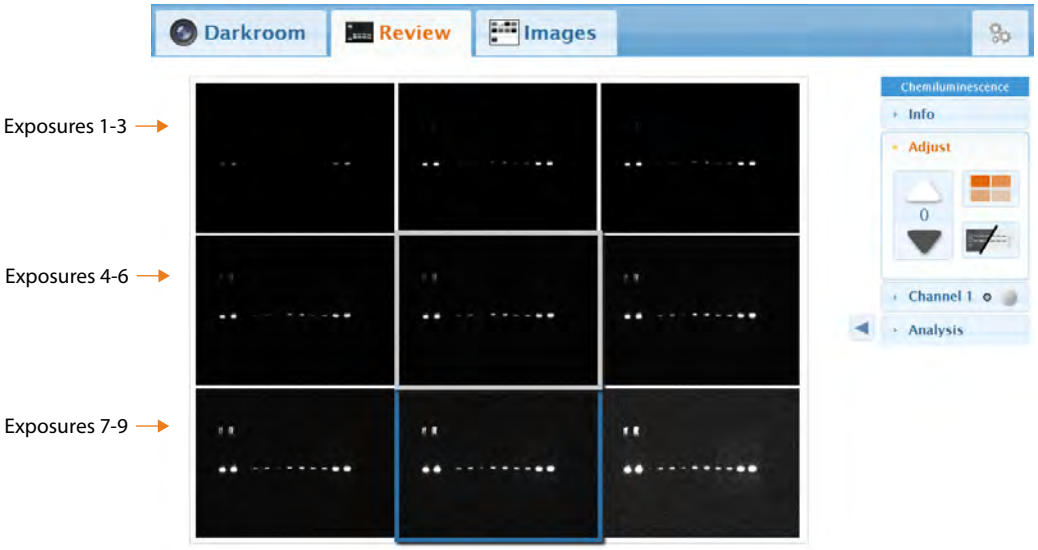


Chemiluminescent Image
After Invert

Exposures View

The Exposures View feature in the **Review Screen** displays additional exposures of the acquired image and allows users to view and select from nine different exposure levels. Once the **Exposures View** button is selected, the following exposures will display:

- **Exposure 1**: Raw image with no exposure corrections.
- **Exposure 2**: Image at optimum contrast with automatic leveling of black and white values so the image is displayed using full gray levels of the screen or monitor. This exposure is selected by default.
- **Exposures 3 - 9**: Display views of increasing exposure levels from Exposure 2.



Select the image with the desired exposure

The current exposure selection will be highlighted in dark blue. The exposure selected in this view is used as the default image display for the **Review** and **Images** screens, and will be the default 8-bit .png image stored with the .fcz file.

To choose a different exposure as the default image, simply select the desired exposure. To exit the page, select the **Exposures View** button again.

Image Zoom

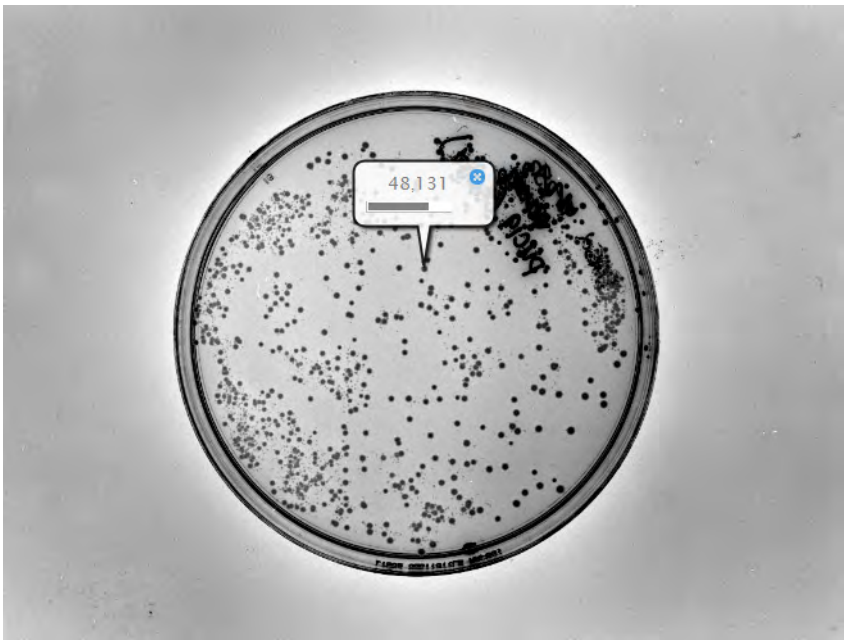
The **Review Screen** also allows users to enlarge the size of the displayed image using the Zoom option. The **Zoom** button can be toggled between **200%**, **400%** and **Off**.

To enlarge an image, toggle the **Zoom** button to the desired magnification level. To view other areas of the enlarged image, select and drag the image until the desired viewing area is shown. To exit zooming, toggle the **Zoom** button to **Off**.

Zoom Off
22 of 28
Delete
Download
Print
Older
Newer
200% Zoom
22 of 28
Delete
Download
Print
Older
Newer
x2
400% Zoom
22 of 28
Delete
Download
Print
Older
Newer
x4

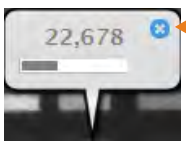
Viewing Pixel Values

Pixel signal intensity can be determined using the Pixel Viewer option. To view pixel values, select the pixel of interest on the image.



The Pixel Viewer will display pixel values as well as a horizontal bar graph indicating the degree of saturation for each channel. Saturation is indicated by a value of 65,636.

NOTE: The saturation value for stacked channels (those that use the same light source) increases by 65,636 per additional stacked channel.



Pixel values for other locations on the image can be viewed by selecting another pixel of interest. To exit the Pixel Viewer, select the **Close** button in the upper right corner of the values box.

NOTES:
Pixel values and degree of saturation depend on the channel image data currently displayed. Channels that are hidden from view are not included in pixel value calculations.

Calculated pixel values are based on 16-bit image files and are unaffected by image adjustment settings.

Browsing Images

The **Older** and **Newer** buttons allow users to browse images saved on FluorChem E in the **Review Screen**. Selecting either button will show images as they are currently displayed in the **Images Screen**.

FluorChem E System File Information

FluorChem E system files have a *.fcz file extension. If a **Title** was entered in the **Image Info** box, this will also be used for the file name. For example, if the title is 'LOD experiment' the file name will be LOD experiment.fcz. If a title is not entered, the default file name will include the date and time of acquisition. An example default file name would be: 2010-06-16_09-22-15.fcz.

The .fcz file format is a proprietary zipped file format that can be opened directly in AlphaView software for post-acquisition data quantitation. Each file contains image acquisition and user-entered information as well as 16-bit .png files of each channel image and an 8-bit per channel .png file. This 8-bit per channel .png file is the overlaid multi-channel image or single channel image based on the selection in the **Exposures** page.

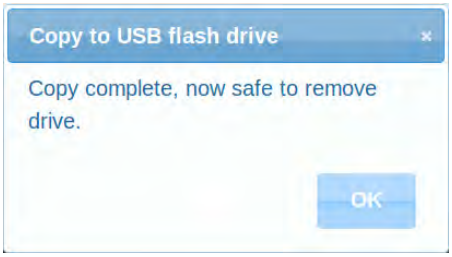
Once opened in AlphaView software, FluorChem E system .fcz files can be converted to .tif or .jpeg files.

Copying, Printing and Deleting Images

Copying Images

To copy an image, select the **Copy** button. Insert a USB flash drive into one of the three USB ports on the FluorChem E system. When the drive is detected, the following message is displayed:

Select the **Copy** button from the dialog box. When the copy is complete the following screen will display:



NOTE: Do not remove the USB drive from the USB port until the copy process has completed or is cancelled.

The **Copy** command will automatically copy a folder to the USB drive. The folder name will be the same as the .fcz file and will contain the following files:

- **FluorChem E system .fcz file**. This file can be used for image analysis in AlphaView software.
- **16-bit .png image file(s)**. These full bitmap files are ideal for quantitation and use in print publications. A 16-bit .png file is provided for each channel.
- **8-bit per channel .png image file**. This smaller sized file is ideal for use in documentation and presentations as well as for e-mailing. This .png file is the overlaid multi-channel image or single channel image based on the selection in the **Exposures** page.

The **Title** information entered by the user in the **Image Info** box will be used for both .png file names. Example .png file names for an image with 2 channels would be: New_Prep_Method_8bit.png, New_Prep_Method_1_16bit.png and New_Prep_Method_2_16bit.png. Channel numbers are appended to 16-bit .png file names.

*NOTE: If **Title** information is not entered, the .fcz file name will be used as the .png file names, for example: 2010-06-16_09-22-15_1_16bit.png. However, ProteinSimple recommends changing the default title to a unique entry as this will help identify archived image files.*

Printing Images

Images saved on the FluorChem E system can be printed to a digital grayscale thermal USB printer. Please contact ProteinSimple Technical Support to obtain information on compatible printers.

To print an image, select the **Print** button. The system will first confirm a printer is available and then display the following dialog box:

Select the **Print** button from the dialog box.

Printouts will contain the title and project entered by the user in the **Image Info** page, as well as the system and protocol information stored with the image:

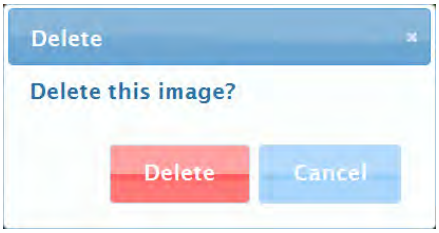
- Protocol settings
- Date and time of image acquisition
- File name
- Instrument name and location

The following is an example of an image printout:

Channel Settings
No Light:1:00m, No Light:30s, No Light:30s
Title
2011-03-01_13-33-06
Notes
Initial data set
FluorChem E FE0232: Laboratory
Instrument Name and Location
2011-03-01 01:33PM
Date and Time
Protein Study
Project
FCE_0232_2011-03-01_13-33-06.fcz
File Name

Deleting Images

To delete an image, select the **Delete** button. The following dialog box will display:



Select the **Delete** button in the dialog box.

---

*NOTE: Images deleted from the **Review Screen** are moved to the **Trash** page and are not permanently deleted. After an image has been deleted, the **Images Screen** will display. For details, please refer to "Trash Page" on page 81.*

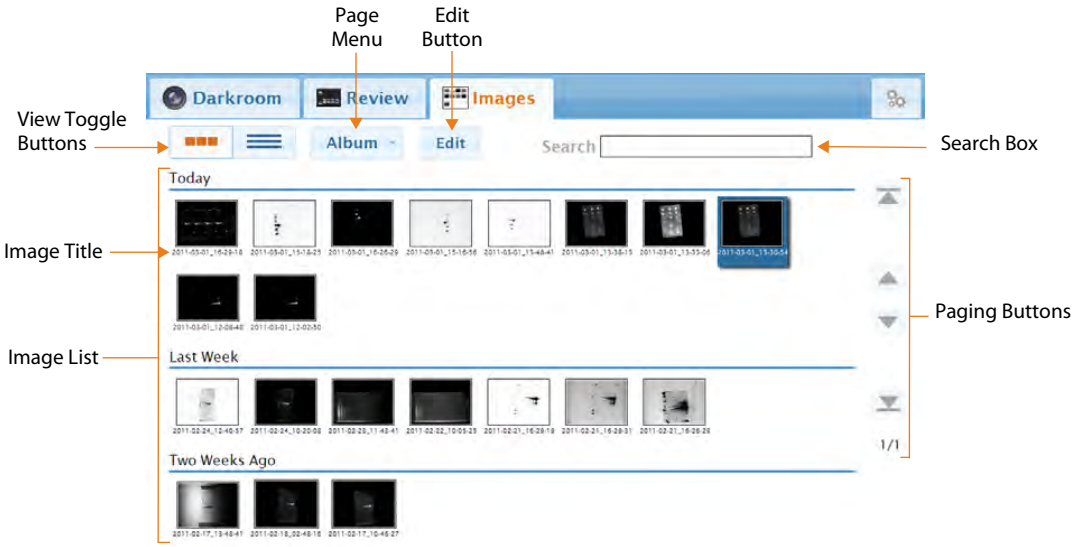
Chapter 7:
Managing Stored Images

Chapter Overview

- Overview of the Images Screen
- Editing the Image Album
- Trash Page

Overview of the Images Screen

Selecting the **Images Tab** will display the **Album** page of the **Images Screen**. This screen is used to browse saved images.



View Toggle Buttons

The **View Toggle** buttons allow users to change how the list of image files is displayed. The view can be toggled between viewing image files as **Thumbnails** or listed in a **Table**.

Image List

The **Image List** displays a complete list of all image files stored on the FluorChem E system. The **Image List** display depends on the **View Toggle** button selected.

Thumbnail View

In the **Thumbnail** view of the **Image List**, file thumbnails are shown for each image. Image files are sorted by date and time, with the most recent files at the top of the list. Image titles are listed below each thumbnail. The **Thumbnail** view will automatically scroll to the image currently displayed in the **Review Screen**, and this image will be highlighted in dark blue.

---

*NOTE: If the **Auto Expose** setting was used and image acquisition was stopped by the user, **Auto Stopped - see protocol details** will display as the image title.*

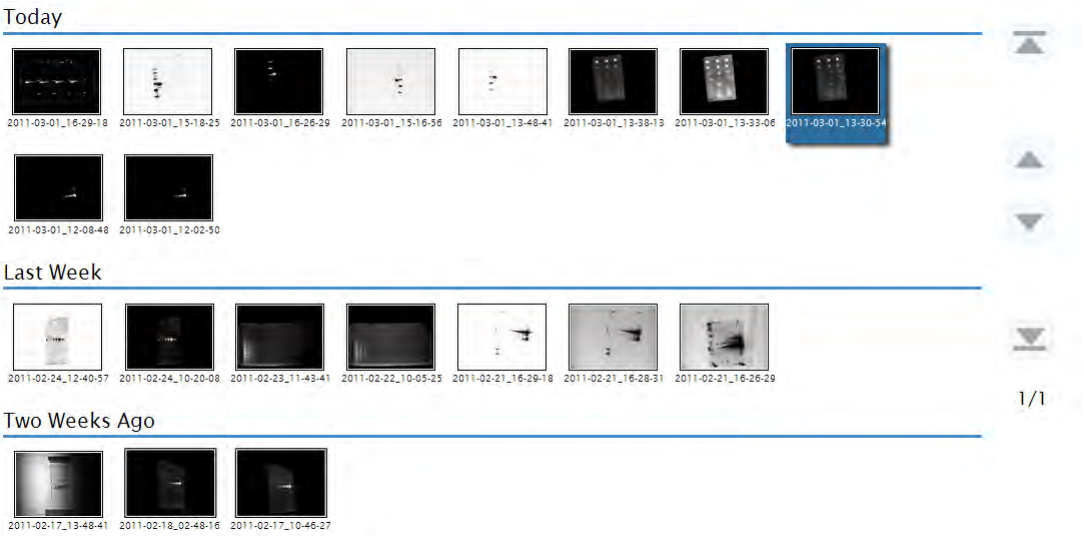


Table View

In the **Table** view of the **Image List**, image files are shown in a table format. Table columns include:

- User, title, project and tags entered by the user
- Acquisition protocol name
- Date and time of image acquisition
- Protocol exposure and resolution settings

NOTES:
***Multiple** will display for exposure settings when image files contain channel data with different exposure times.*

*NOTE: If the **Auto Expose** setting was used and image acquisition was stopped by the user, **Auto Stopped - see protocol details** will display as the image title.*

User	Title	Project	Tags	Protocol	Date	Exposure	Resolution
	2011-03-01_15-16-56			Ethidium Bromide	03/01/2011 3:16PM	270ms	Standard
	2011-03-01_13-48-41			Ethidium Bromide	03/01/2011 2:35PM	630ms	Standard
	2011-03-01_13-38-13			Chemiluminescence	03/01/2011 1:38PM	40s	Standard
PC	2011-03-01_13-33-06	Protein Study	ERK2	Chemiluminescence	03/01/2011 1:33PM	Multiple	Standard
PC	2011-03-01_13-30-54	Protein Study	ERK1	Chemiluminescence	03/01/2011 1:30PM	Multiple	Standard
PC	2011-03-01_12-08-48	Protein Study	ERK2	Chemiluminescence	03/01/2011 12:08PM	Multiple	Standard
PC	2011-03-01_12-02-50	Protein Study	ERK1	Chemiluminescence	03/01/2011 12:02PM	10s	Standard
	2011-02-24_12-40-57			Ethidium Bromide	02/24/2011 12:40PM	920ms	Standard
	2011-02-24_10-20-08			Chemi with Markers	02/24/2011 10:20AM	Multiple	Standard
	2011-02-23_11-43-41			Coomassie Blue	02/23/2011 11:43AM	670ms	Standard
	2011-02-22_10-05-25			Coomassie Blue	02/22/2011 10:05AM	Multiple	Standard
	2011-02-21_16-29-18			Coomassie Blue	02/21/2011 4:29PM	560ms	Standard

By default, image files in the **Table** view are sorted by date, with the most recent files at the top of the list. The **Table** view will automatically scroll to the image currently displayed in the **Review Screen**, and this image will be highlighted in dark blue.

Paging Buttons

The **Paging** buttons allow the user to page through data in the **Image List**. The **Page Up** and **Down** buttons will scroll the **Image List** to the next or previous page of files. The **First** and **Last Page** buttons will scroll the **Image List** to the first or last page of files.

*NOTE: When the **Image List** is at the first or last page of stored image files, the **Paging** buttons will become inactive.*

Page Menu

This menu allows users to switch between the **Album** and **Trash** pages. All images are stored in the **Album** page by default. When an image file is deleted in the **Review Screen** or from the **Album Edit** page, it is moved to the **Trash** page. To view image files in either page, select the **Page** menu.

Search Box

The **Search** box allows users to enter text details to search for specific images.

Edit Button

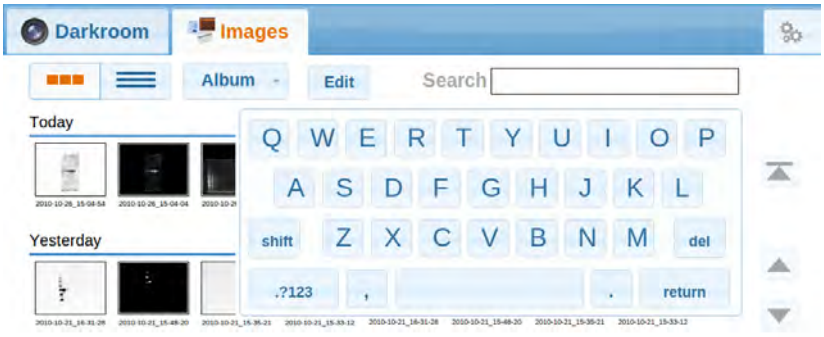
The **Edit** button displays the **Edit** pages where image files can be copied, deleted or moved from the **Trash** page back to the **Album** page.

Selecting Images for Review or Enhancement

To select an image for review or enhancement, select a thumbnail or row in the **Image List**. The selected image will then display in the **Review Tab**. For more information on the **Review Screen** and making image adjustments, please refer to Chapter 6, *"Image Review"*.

Searching for Images

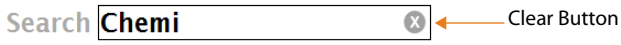
The **Search** box allows users to search for specific images in the Image Album. To search for an image, select the **Search** box. A keyboard will display:



Type in the text search information. The following information can be used:

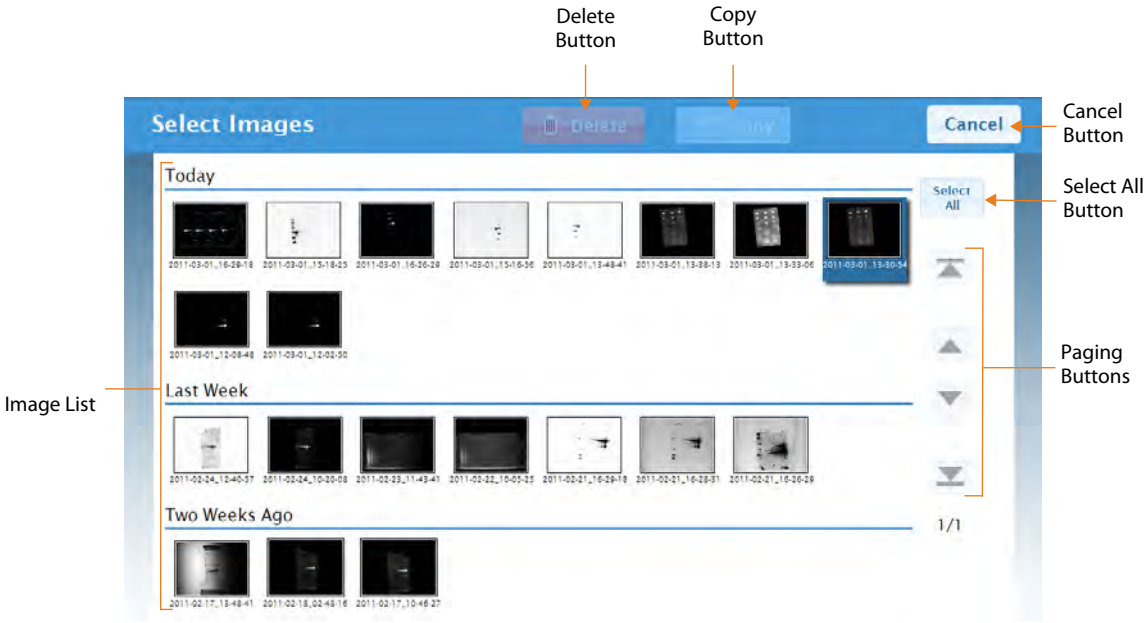
- User entered information: title, user, project, and tags
- Image file information: date, protocol, exposure and resolution

When text entry is complete, select the **Return** button. Search results will be returned in the **Album** page. To clear the text in the **Search** box and return to a view of all files, select the **Clear** button.



Editing the Image Album

To delete or copy images in the Image Album, select the **Edit** button. This will display the **Album Edit** page. The page will automatically scroll to the image currently displayed in the **Review Screen**, and this image will be highlighted in dark blue.



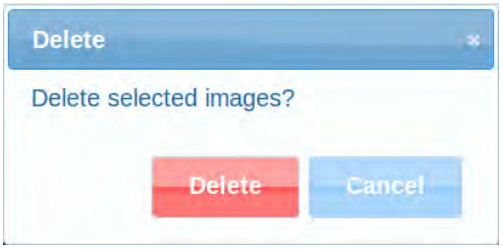
Selecting Images in the Album Edit Page

One or multiple images can be selected in the **Album Edit** page for subsequent deletion or copy. To select images, select the thumbnail(s) or row(s) of the desired images in the **Image List**. Once images are selected, the **Delete** and **Copy** buttons will become active. Selected images will be designated with a check mark, and the number of images selected will display in the upper left corner of the page:

The **Select All** button can be used to select all images on the current page. Selecting the button again deselects all images on the page.

Deleting Images from the Album

To delete one or multiple images from the Image Album, select the image(s) in the **Image List**, then select the **Delete** button. The following dialog box will display:



Select the **Delete** button in the dialog box.

---

*NOTE: Images deleted on the **Album Edit** page are moved to the **Trash** page and are not permanently deleted.*

Copying Images from the Album

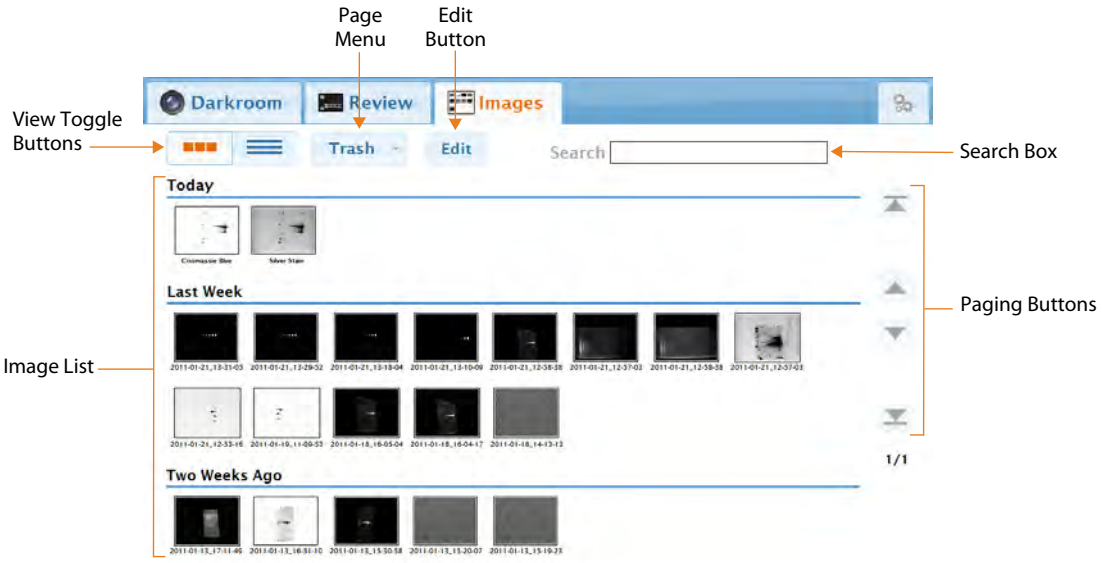
To copy one or multiple images from the Image Album, select the image(s) in the **Image List**. The copy function in the **Album Edit** page is the same as described in "Copying Images" on page 67 except that multiple folders are copied to the USB drive.

Exiting the Album Edit Page

To exit the **Album Edit** page, select the **Cancel** button.

Trash Page

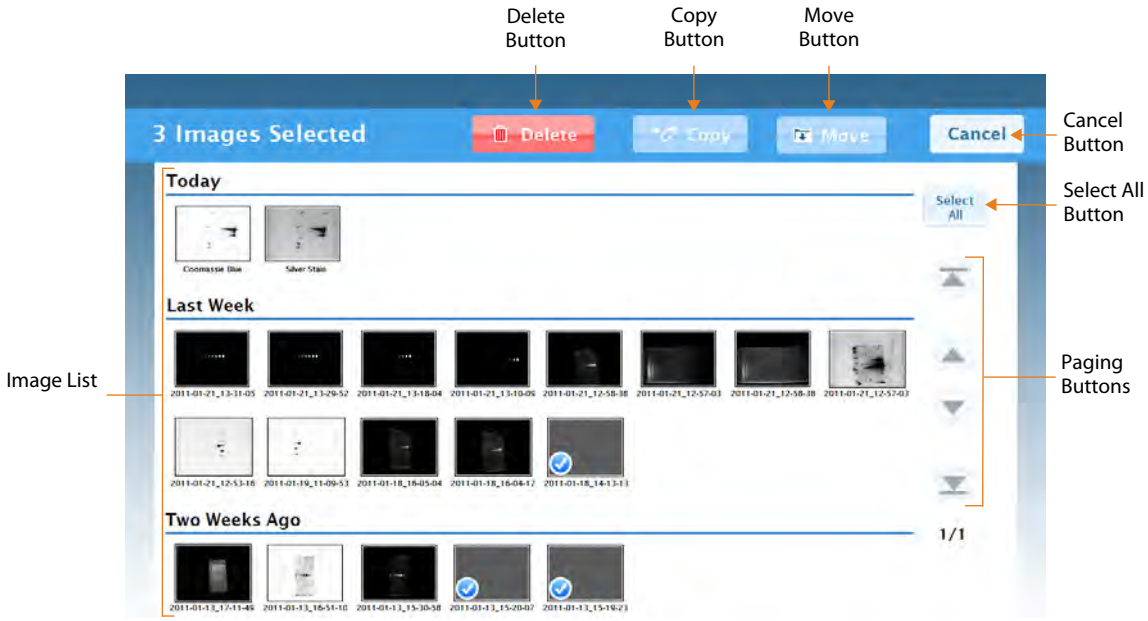
The **Trash** page contains images that have been deleted from the **Review Screen** and **Album Edit** pages. Users can search and browse through all stored image files in the this page. To view the **Trash** page, select it from the **Page Menu**.



Trash page options are the same as the **Album** page. Please refer to "Overview of the Images Screen" on page 74 for details on how to use this page.

Editing the Trash

To delete, copy or move images in this page, select the **Edit** button. This will display the **Trash Edit** page.



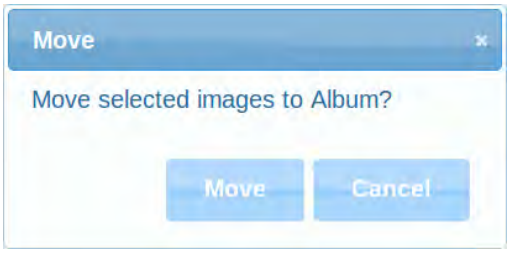
With the exception of how images are deleted and the **Move** button, the **Trash Edit** page options are the same as the **Album Edit** page. Please refer to "Editing the Image Album" on page 79 for details on how to use this page.

Deleting Images from the Trash

Image files can be deleted from the **Trash Edit** page in the same way that they are deleted in the **Album Edit** page. However, images deleted from the **Trash Edit** page are permanently deleted.

Moving Images back to the Image Album

To move one or multiple images back to the Image Album, select the image(s) in the **Image List**, then select the **Move** button. The following dialog box will display:



Select the **Move** button in the dialog box. The image file(s) will be moved out of the Trash and back into the Image Album.

Exiting Trash Selection

To exit the **Trash Edit** page, select the **Cancel** button.

Backup and Restore

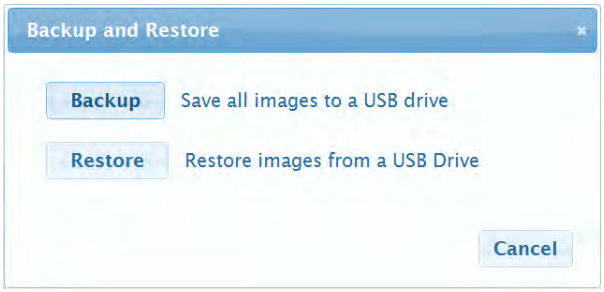
Backup

Acquired images are stored on the internal hard drive of the imager. It is recommended that images be backed up regularly to an external USB flash drive.

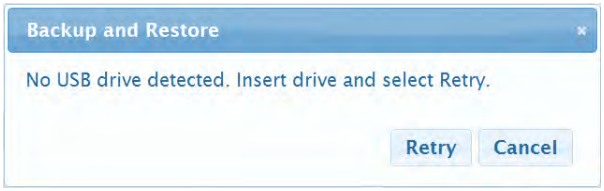
NOTE: Backup and Restore is not visible on the remote browser page, only at the instrument.

To initiate the backup, select **Backup** on the **Settings** screen.

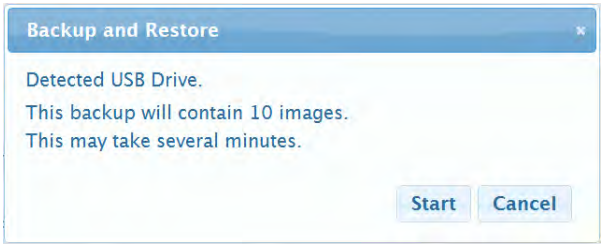
After selecting **Backup**, it will open a dialog giving you the choice to initiate a **Backup** or **Restore**. Restoring images from a USB will not overwrite existing files on the imager.



The following dialog box appears if no USB is detected:

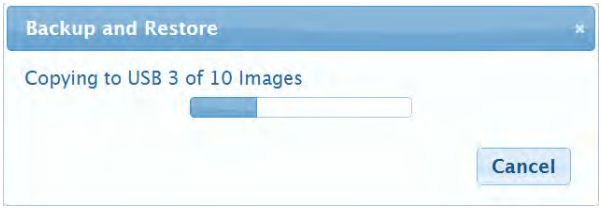


Backup displays a dialog box containing info on the backup before it starts.



If the USB drive does not have room for the backup, a warning is given with the amount of room required.

During the backup, the status appears:



When backup is completed, the following dialog box appears:

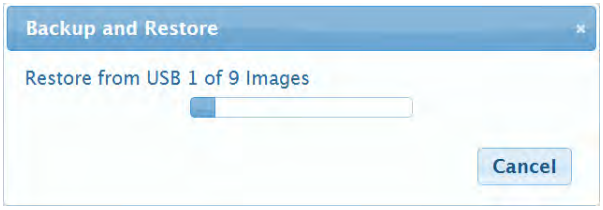
Restore

Insert the USB containing backup files. The instrument will search the USB drive for the backup folders, but will only show the latest backup. To restore from an earlier backup, the more recent backups need to be removed from the USB.

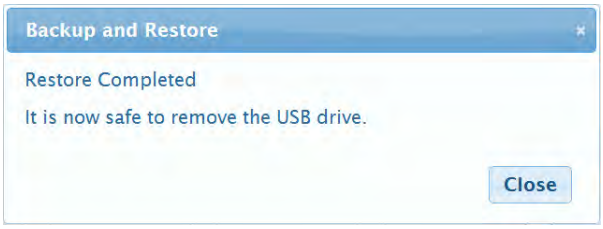
The **Start Restore** dialog box reports how many backed up images the folder contains. The term *images* refers to *runs* which may have multiple images each.

All existing runs are maintained during the restore. If a backed up run already exists on the instrument it is skipped in the restore, so changes to a run since the backup will be maintained. If a run restored to the album page is also in the trash, the copy in the trash is deleted.

During restore, the status appears:



When restore is completed, the following dialog box appears:



Chapter 8:

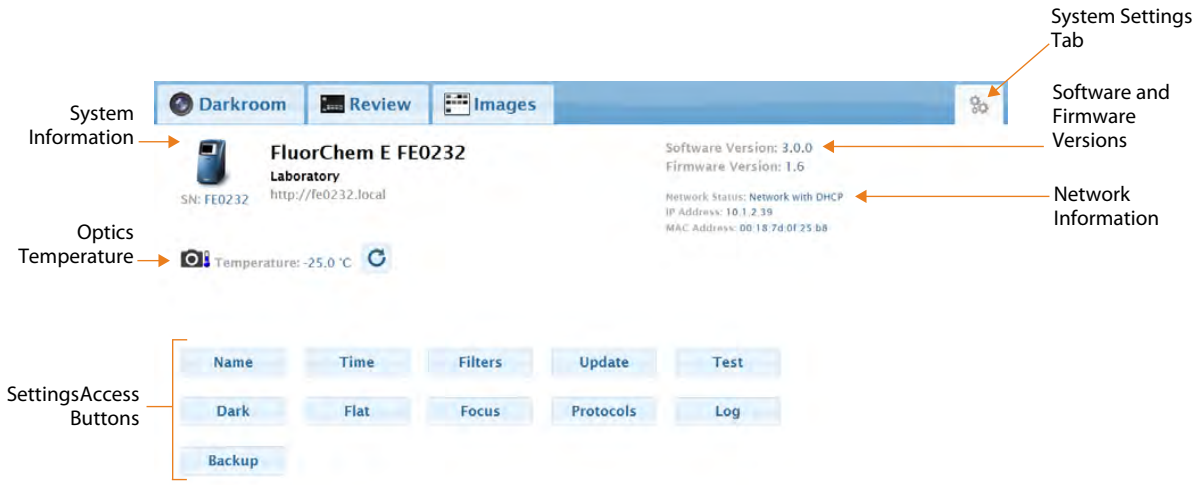
System Settings

Chapter Overview

- Overview of the System Settings Screen
- Changing System Information
- Changing Date and Time Settings
- Changing Filter Settings
- Updating the Software
- Saving Custom Protocols
- System Self Test
- Creating Dark Masters
- Flat Field Calibration
- Changing Focus Settings
- System Log

Overview of the System Settings Screen

Selecting the Settings Tab will display the System Settings Screen. This screen contains system information and allows users to change the system name, date and time and focus settings as well as install software updates and run system tests. Dark masters, flat field calibration and system log files can also be generated.



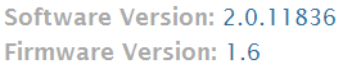
System Information

FluorChem E system information is located in the upper left corner of the System Settings Screen.

The system serial number is located under the instrument icon in blue text. The FluorChem E system name and location are included on image printouts and are also used to identify the instrument on the network. System name and location information can be customized by the user by clicking on the Name button. The default system name will include the serial number (for example FE0232).

Software and Network Information

The current versions of the FluorChem E Digital Darkroom software and firmware are displayed in the upper right corner of the screen.



Network information is located just below the version information and displays the FluorChem E system network status, IP and MAC addresses.

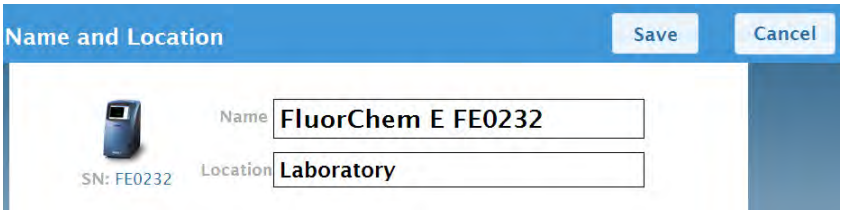
NOTE: If the FluorChem E system is not connected to a local network, the Network Status will display 'local connection (no network)'.

Optics Temperature

The current temperature of the FluorChem E system optics is also shown in the System Settings Screen. The temperature displayed is a static measurement taken at the time the screen is accessed. To get an updated reading at any time, select the Refresh button.

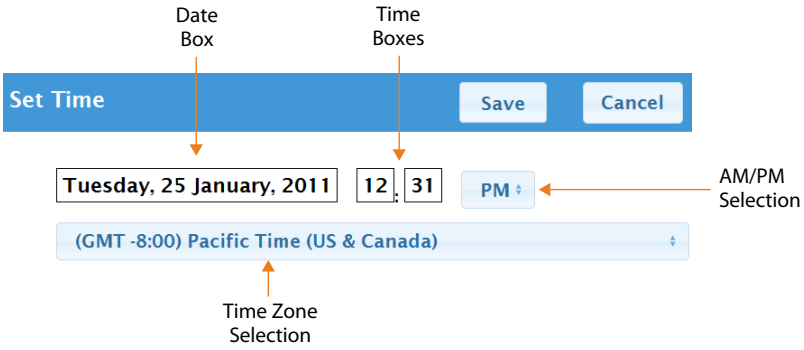
Changing System Information

To change the FluorChem E system name or location setting, select the Name button. Select either text box and use the keyboard to enter the new information. When text entry is complete, select the Save button.



Changing Date and Time Settings

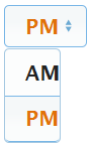
To change the date, time or time zone setting, select the Time button. The following settings will display:



To change the date, select the Date box and choose a new date from the calendar provided. The current date will be displayed in yellow. Use the arrows at the upper left and right to change the calendar month.

To change the time, select hours or minutes in the Time boxes and use the number pad to enter the new information.

The AM or PM setting can be changed by selecting the AM/PM menu box. The current setting will be displayed in orange text.

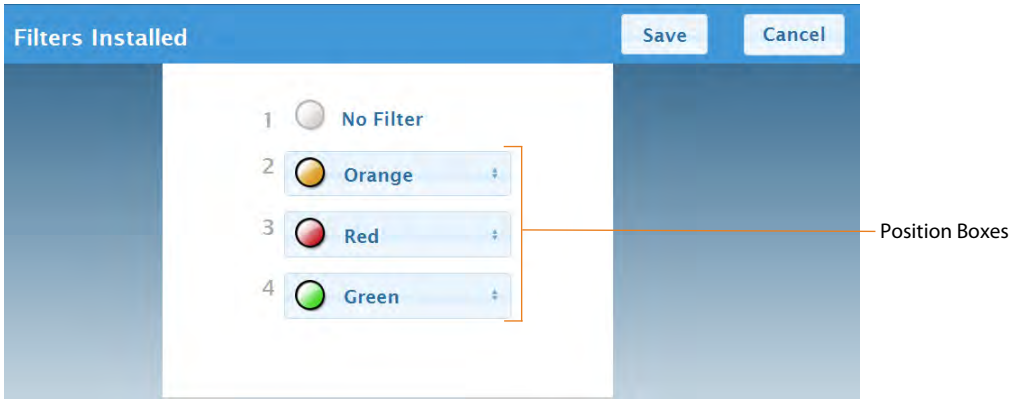


To change the time zone, select the Time Zone menu box and choose an option from the drop down list. The current setting will be displayed in orange text.

When the desired date and time settings have been updated, select the Save button to save the changes.

Changing Filter Settings

To change filter settings, select the Filter button. This will display the type of filters currently used in the FluorChem E system and their locations in the filter wheel.



Position 1 does not contain a filter and is used whenever a protocol uses the No Filter option. The Position 1 setting cannot be changed by the user. Positions 2-4 can be set by the user and should reflect what filters are currently installed in the filter wheel.

To change the filter setting for a particular filter position, select a Position box and choose a filter from the drop down list.

If the position does not contain a filter, the setting should be set to Empty. Filter options for each position include orange, red, blue, green or empty. When filter settings have been updated, select the Save button to save the changes.

Please refer to "Installing Emission Filters" on page 116 for instructions on installing filters in the filter wheel.

Updating the Software

Software updates can be transferred to the FluorChem E system using a USB flash drive or downloaded via the web.

USB Updates

1. Insert the USB drive into any USB port on the instrument.
2. Select the Update button. The following dialog box will display:

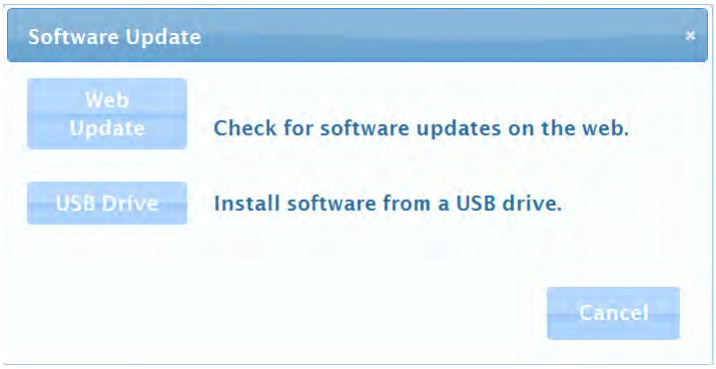
3. Select the USB Drive button.
4. The software will perform an automatic scan of the USB drive to locate the software installer file. When prompted, follow the on-screen instructions to complete the software update.

Web Updates

NOTE: Web updates require that the FluorChem E system be connected to a local network with internet access.

---

1. Select the Update button. The following dialog box will display:

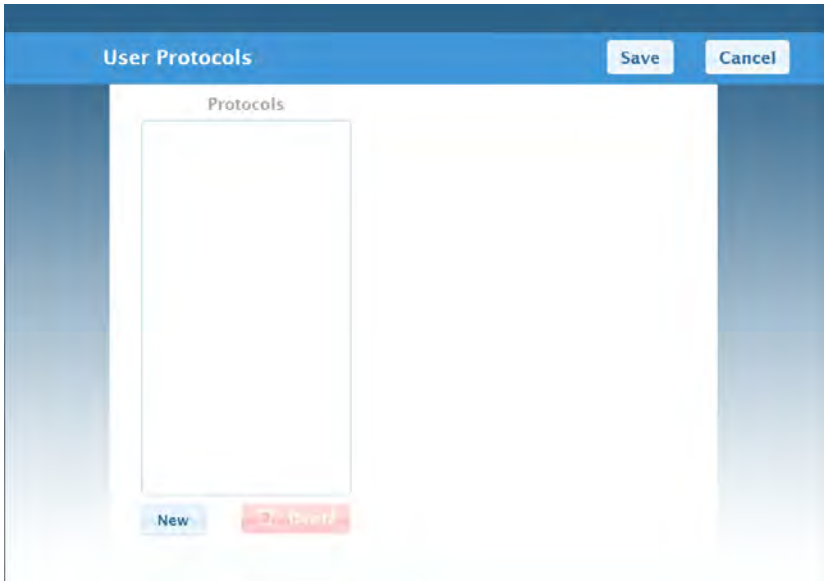


2. Select the Web Update button.
3. The software will automatically check to see if a new version of the software is available. If one is found, follow the on-screen instructions to complete the software update.

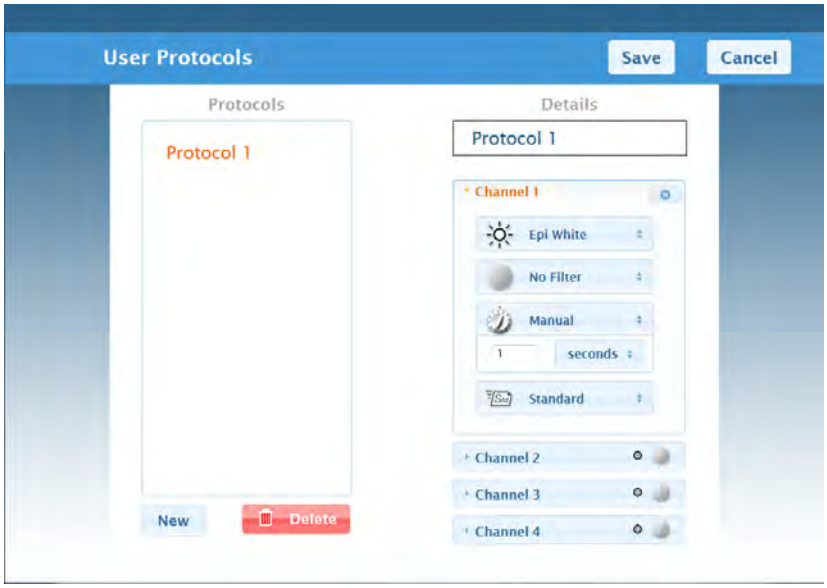
Saving Custom Protocols

Custom protocols can be created and stored for future use. These can be accessed through the drop down protocol menu (see page 38).

1. To create a protocol, select Protocols. The User Protocols screen allows for both the creation of new protocols and modification of existing protocols. Factory-supplied protocols cannot be edited.



2. Select New to bring up the protocol details. The protocol name and read settings can be entered here.



3. Click Save to save the User Protocol or Cancel to discard changes.
4. Protocols can be deleted by selecting the protocol and then Delete.

System Self Test

FluorChem E system performance can be tested to ensure key components are functioning properly. To run the system self tests or view results from previous tests, select the Test button.

A Test Log of previously run self tests with overall pass/fail results is shown on the left. To view results for a particular test, select the desired Test Date in the log. The Results Table will display the start time, individual test result and failure reason for each component tested.

Running the Self Tests

1. Open the FluorChem E system door and remove any sample screens from the UV transilluminator.
2. Close the system door.
3. Select the Run Test button. FluorChem E will automatically perform individual component tests and report results. Testing takes approximately 5 minutes to complete.

To copy test file results, insert a USB drive into any USB port on the instrument and select the Copy button.

NOTE: If any of the tests return a failed result, please contact ProteinSimple Technical Support.

Creating Dark Masters

NOTES:
Dark masters are generated on the FluorChem E prior to system shipment. Dark Master files typically only need to be updated as required for system service.

Dark Master file generation requires approximately 12 hours of uninterrupted system time, during which the FluorChem E may not be used for image acquisition. ProteinSimple recommends starting the process in the evening and letting it run overnight.

---

1. Close the FluorChem E system door.
2. Go to the System Settings Tab and select the Dark button. The following dialog box will display:

3. Select Start. This following status screen will display:

The start and expected completion times will be shown, along with an image counter and progress bar which will update during the Dark Master file generation process.

NOTE: To maintain optimal light-tight conditions, avoid opening the FluorChem E system door after starting Dark Master file generation.

---

4. When the Dark Master files have been created, the following message will display to indicate the process is complete:

Click OK to exit.

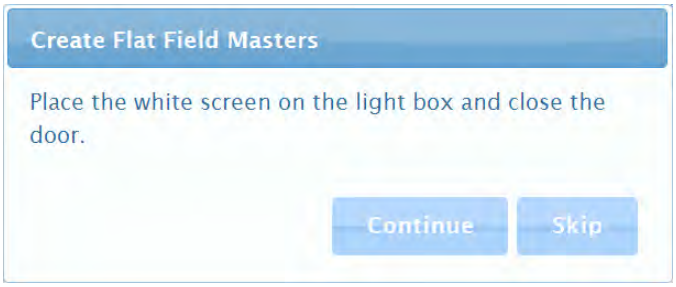
Flat Field Calibration

Flat field calibration is used to correct for optical non-uniformity inherent in CCD based imaging systems. This calibration is applied automatically as part of the image acquisition process.

IMPORTANT: Flat field calibration is performed on the FluorChem E prior to system shipment. Recalibration is periodically recommended depending on variations in illumination across the field of view. Please contact ProteinSimple Technical Support for further assistance.

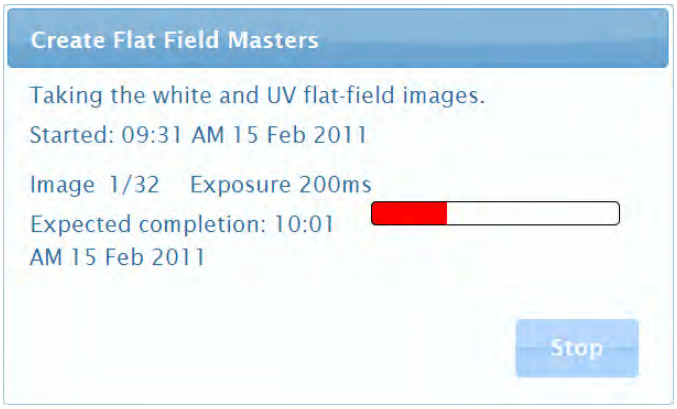
1. Go to the System Settings Tab and select the Flat button. The following dialog box will display:

2. Select Start. This will prompt the following message:



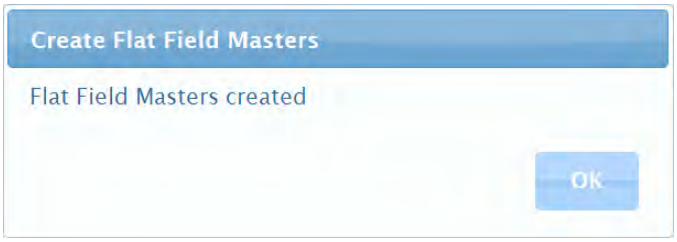
If UV/white light flat field calibration is not required, select Skip to proceed to the completion step. Otherwise, place the **white screen used for colorimetric imaging** on the UV transilluminator.

Close the system door and select Continue. The following status screen will display:



The start and expected completion times will be shown, along with an image counter and progress bar which will update during the calibration process.

3. When the UV/white light flat field calibration has finished or was skipped, the following message will display to indicate the process is complete:



Click OK to exit.

Changing Focus Settings

NOTE: The FluorChem E system optics are set to an optimal fixed focus above the sample screen and do not normally need to be changed. However, if the focus settings do need to be adjusted, use the following procedure.

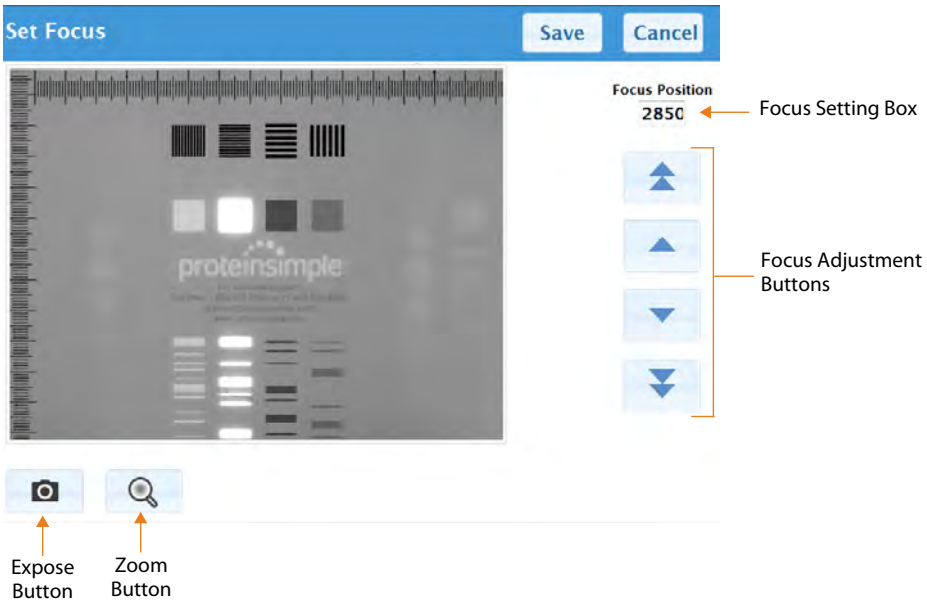
1. Place the imaging test target provided with the FluorChem E system on the black sample screen and center the target on the imaging area.
2. Place the screen on the UV transilluminator.

3. Close the system door. A white light image of the target will automatically be taken and displayed.
4. Go to the System Settings Tab and select the Focus button. The Focus Screen will appear and the system will automatically take and display new white light image.

NOTE: White light focus images may be blurry until optimum focus has been reached.

---

5. Adjust the focus incrementally by selecting the Focus Adjustment buttons until the optimum focus is reached. The two middle buttons (single arrows) will adjust the focus up or down in minor increments. The top and bottom buttons (double arrows) will adjust the focus up or down in major increments. Alternatively, users can also enter a setting directly in the Focus Setting box.



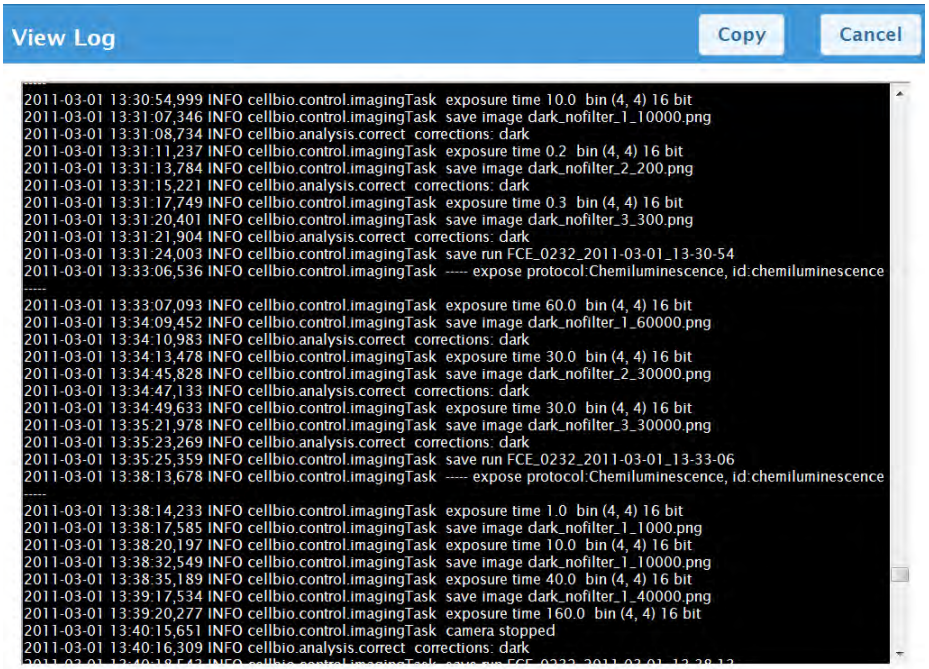
As adjustments are made, the value in the Focus Setting box will update automatically. A new white light image will be taken and displayed each time the focus is adjusted.

Selecting the Expose button will take an additional epi-white light image. The Zoom button can be used to magnify white light images as desired.

6. When the desired focus is reached, select Save to save the new setting. The Focus Screen will close and the System Settings Screen will be displayed.
7. Select the Darkroom Tab. A new white light image will be taken and displayed using the revised focus setting.

System Log

To view or copy a FluorChem E system log file, select the Log button. The following screen will be displayed:



Use the scroll bar to review the log in its entirety. To copy the log file, insert a USB drive into any USB port on the instrument and select the Copy button.

Chapter 9:

Networking and Remote Access

Chapter Overview

- Digital Darkroom Remote Access
- Connecting to FluorChem E
- Downloading Image Files
- Opening Image Files in AlphaView Software

Digital Darkroom Remote Access

If FluorChem E is connected to a local network, image acquisition can be monitored and controlled directly from any networked computer or mobile Wi-Fi device through a web browser. Stored image files can also be accessed remotely for reviewing, copying, printing and desktop image analysis using AlphaView software.

Connecting to FluorChem E

FluorChem E can be accessed remotely via a permanent URL address or a dynamic IP address.

NOTE: The FluorChem E URL address is a unique and permanent address and will not change. The IP address, however, is not permanent and can change if the system is ever disconnected and reconnected to the local network.

FluorChem remote access via URL requires Apple® Bonjour (zero-configuration networking) to be installed on Windows®-based computers and devices. Bonjour can be downloaded at http://www.apple.com/support/bonjour. Download and install **Bonjour Print Services for Windows**.

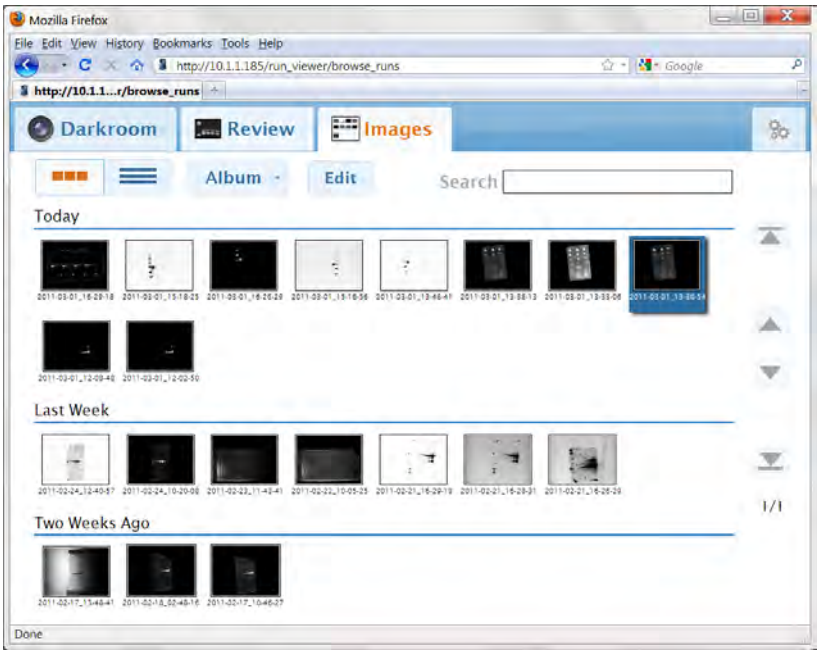
NOTES:
Once installed, it is not necessary to launch Bonjour prior to accessing the FluorChem E system remotely.

Bonjour capability comes pre-installed on Mac, iPad, and iPhone and does not need to be installed separately for remote web access of the FluorChem E system.

To connect to a networked FluorChem E system:

1. Open a web browser on a computer or device that is on the same network as the FluorChem E system.
2. Enter the URL or the current IP address for the FluorChem E system in the browser window. The system's URL and IP address information can be found in the **System Settings Screen.**

3. The browser will expand the entered URL or IP address to http://feXXXX.local/run_viewer/browse_runs/ or http://xx.x.x.xxx/run_viewer/browse_runs/ and display the **Images Screen**.



4. The full FluorChem E Digital Darkroom remote interface will now be accessible.

*NOTE: If FluorChem E is executing a calibration or self test when accessed remotely, an **Instrument Test** message will display in the **Darkroom Tab**. If this occurs, data can still be accessed from the **Images** and **Review** tabs. Image acquisition and monitoring in the **Darkroom Tab** will remain unavailable until the calibration or test is complete.*

Using the remote access interface is similar to using the touchscreen interface directly on the FluorChem E system except for the following functions:

- **Text entry**. The keyboard on the remote computer or device is used for all text and numeric entry.
- **Printing**. The **Print** command will print image files to a local or network printer only.
- **Copying Image Files**. In the remote interface, an option to download files is provided instead of the copy option.

Downloading Image Files

Stored image files can be downloaded to the remote computer or device. Users are provided the option to either download the .fcz file or just the .png files.

NOTE: Depending on the option selected, downloaded files can be provided in .zip format. WinZip® or a similar extraction program is required to extract .zip files.

To download files:

1. Select the **Download** button in the **Review Screen** or from an **Edit** page in the **Images Screen**.

*NOTE: Multiple images can be selected for download in the **Album Edit** or **Trash Edit** pages in the **Images Screen**.*

1. The following dialog box will display:

2. Choose a download option:
   - Select the **AlphaView** button to download the .fcz file. If desired, this option can launch AlphaView software automatically after download.
   - Select the **Images** button to download only the 8 and 16-bit .png image files.

NOTES:
*If more than one image is selected when using the **AlphaView** download option, a single zip file will be downloaded that contains all the .fcz files for the selected images. If only one image is selected, a single .fcz file will download.*

*When using the **Images** option, all .png files for an image are provided in a single .zip file.*

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3. Follow the instructions in the browser dialog box to download the file(s).
   - **Single .fcz file downloads.** The .fcz file can either be saved to the remote computer or device or AlphaView software can open it automatically.
   - **Zip file downloads.** Save the .zip file to the remote computer or device. Then extract the file to the desired location using WinZip or other extraction program.

NOTES:
To open .fcz files, AlphaView Stand Alone software version 3.3 or higher must be installed on the remote computer.

The remote computer must be Windows-based to run AlphaView software.

Opening Image Files in AlphaView Software

Stored image files can be opened directly in AlphaView software for post-acquisition image analysis.

NOTES:
To open .fcz files, AlphaView Stand Alone software version 3.3 or higher must be installed on the remote computer.

AlphaView software is only available in a Windows-compatible version.

Both .fcz and .png files can be opened in AlphaView software.

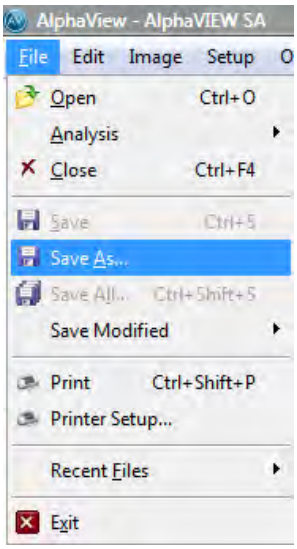
Opening .fcz Files

- **Opening files during download.** Follow the instructions in the browser dialog box to download the .fcz file. When download is complete, AlphaView software will automatically launch and open the file.

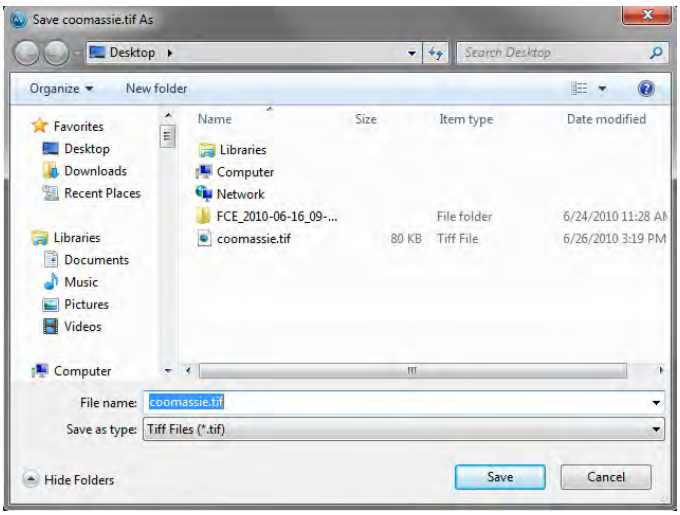
NOTE: Some browsers may prompt for the location of the AlphaView software .exe file on the remote computer. If this occurs, select the Browse option in the browser dialog box. Browse to where AlphaView software is installed (such as Program Files\ProteinSimple\AlphaVIEW SA) and select the AIC.AlphaView.exe file.

- **Opening saved or extracted files on the remote computer.** This can be done in one of two ways:
  - Browse to the folder where the file is located and double-click the desired .fcz file to automatically launch AlphaView software and open the file.
  - Open AlphaView software. Select **File** > **Open**. Browse to the file location and select the desired .fcz file.

In AlphaView software, the displayed *.fcz file can be renamed and saved as a 16-bit .tif file. To do this, select **Save As** from the **File** menu.



The following dialog box will display. In the **Save as type** drop down menu, select **Tiff Files (*.tif)**. The original file name can be changed if desired.



Opening .png Files

To open saved or extracted .png files on the remote computer, open AlphaView software. Select **File** > **Open**. Browse to the file location and select the desired .png file.

NOTE: FluorChem E .png image files can also be opened in any other program that accepts files in .png format.

Chapter 10:
Maintenance and Troubleshooting

Chapter Overview

- Maintenance
- Accessories and Spare Parts
- Troubleshooting

Maintenance

Cleaning the FluorChem E System

Instrument and Sample Screens

Items needed: Cleaning solution (general purpose glass cleaner, isopropanol or 70% ethanol) and a soft cloth or paper towels

1. Saturate a paper towel or cloth with cleaning solution.
2. Wipe down the surface of the UV transilluminator, sample screens and the instrument's exterior blue housing.

LCD Touchscreen

Items needed: LCD screen cleaner (Klear or iKlear) and a microfiber cloth

1. Follow the instructions provided on the LCD screen cleaner bottle.

NOTE: Never spray cleaning solution directly onto the LCD touchscreen.

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2. **If there are no instructions on the cleaner bottle:** Spray the cleaner onto the cloth and gently wipe the screen in a circular motion. Do not press hard while cleaning the screen. Continue to wipe until all of the solution is removed and the screen is completely dry.

NOTE: Leaving excess cleaning solution on the LCD touchscreen may cause long-term damage to the screen.

System Decontamination

The following decontamination procedure must be performed should the FluorChem E system come in contact with hazardous samples.

Items needed: 10% bleach solution and a soft cloth or paper towels

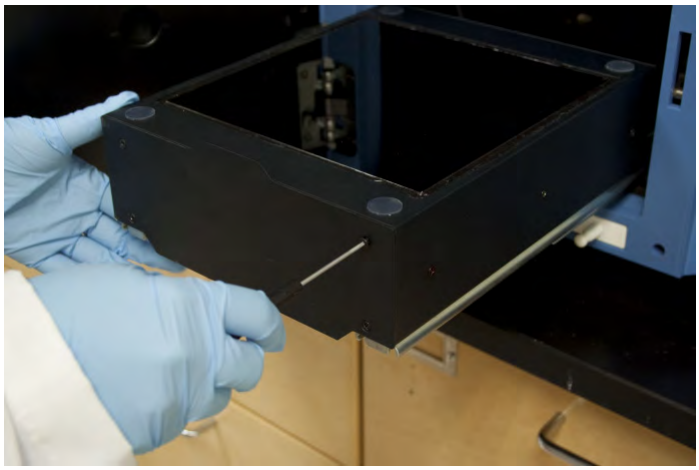
1. Wear rubber gloves.
2. Completely saturate a paper towel or cloth with 10% bleach solution.

3. Wipe down the surface of the UV transilluminator, sample screens and any other areas inside or outside the system that may have come into contact with hazardous samples.
4. Wipe down the exterior of the instrument.

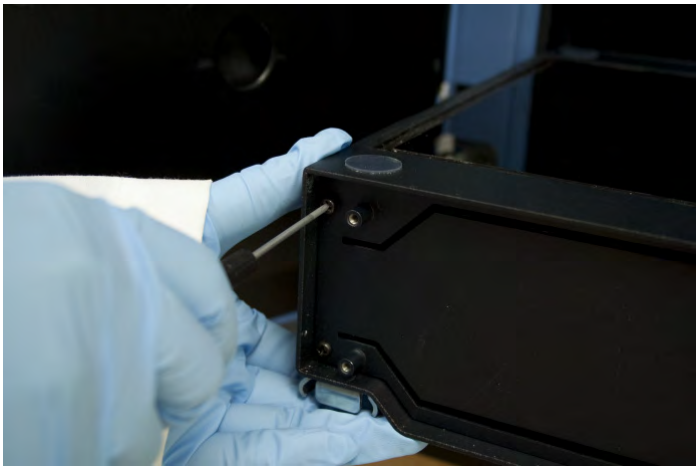
UV Transilluminator Bulb Replacement

NOTE: To ensure uniform illumination, ProteinSimple recommends replacing all six UV transilluminator bulbs at the same time.

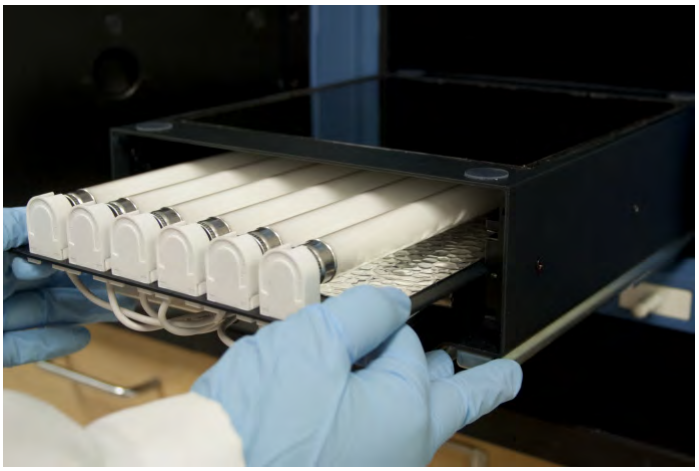
1. Power down the FluorChem E system and unplug the power cord from the system.
2. Open the system door and slide out the UV transilluminator approximately halfway.
3. Using a Philips head screwdriver, unscrew the four screws on the UV transilluminator outer front panel and remove the panel. This will expose the inner front panel.



4. Next, unscrew the four screws on the UV transilluminator inner front panel and remove the panel.



5. Slowly slide out the bulb tray.



NOTE: Remove the bulb tray gently. The bulb tray electrical cord is hard-wired directly to the UV transilluminator and the cord cannot be disconnected. Pulling or tugging on the bulb tray may damage the cord and lead to malfunction in the UV transilluminator.

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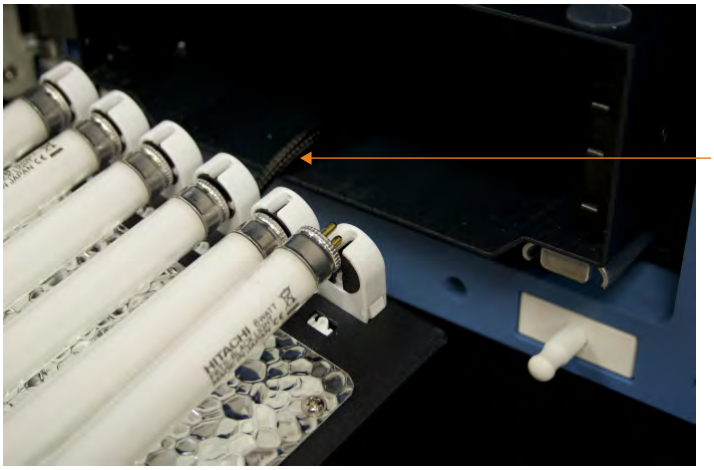
6. To remove a bulb, rotate it until the pins align with the slot in the bulb socket. Next, lift the bulb up and out of the socket.



7. To insert a bulb, slide the pins into the bulb socket and then rotate the bulb a quarter turn. A faint click will be heard when the bulb is seated correctly.

8. Repeat steps 6 and 7 for the remaining five bulbs.
9. Align the bulb tray on the middle notch of the UV transilluminator housing and slide the tray all the way back into the housing.
10. Reinstall the inner front panel and tighten the four screws until snug.
11. Reinstall the outer front panel and tighten the four screws until snug.

12. Push the UV transilluminator back into the system.
13. Plug the FluorChem E power cord back into the wall socket and turn the system power on.

Installing Emission Filters

1. Power down the FluorChem E system.
2. Open the system door and locate the panel in front of the filter wheel at the rear of the inside cabinet.

3. Using a Philips head screwdriver, unscrew the three screws on the base of the panel and remove the panel. This will expose the filter wheel. Each filter position is labeled with a position number.

NOTE: Filter Position 1 is reserved for chemiluminescent imaging and should always remain empty.

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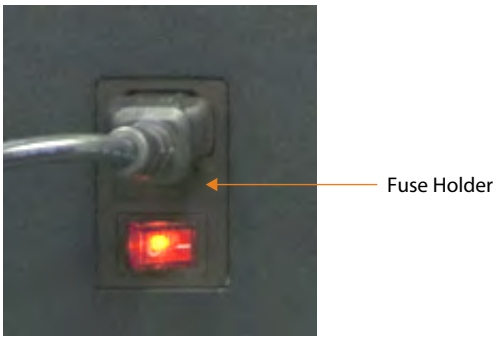
4. To install a filter, insert the threaded side into an open port on the filter wheel. Turn clockwise to screw the filter in place until snug.

5. To remove a filter, grasp the outer edge and turn counter clockwise to unscrew and remove.
6. When filter installation is complete replace the panel.
7. Turn the FluorChem E system power on.
8. Update the Filter Settings in the **System Settings Screen**. For details on how do to this, please refer to "Changing Filter Settings" on page 91.

Changing the Fuse

1. Power down the FluorChem E system and unplug the power cord from the system.
2. Locate the fuse holder on the rear panel of the instrument:



3. Use a flat-head screwdriver to gently pry the fuse holder open. Remove the fuse holder.
4. Remove both fuses and replace them with two 250 V, 6 A fuses (P/N 011-597).
5. Reinsert the fuse holder.
6. Plug the FluorChem E power cord back into the system and turn the power on.

Accessories and Spare Parts

A set of common spare parts, accessories and service information is listed below. To place an order, please call (888) 607-9692.

Part Description	Part Number
Accessories	
Chemiluminescence Imaging Screen (Black)	90-15045-00
UV Gel Imaging Screen (Clear)	90-15044-00
White Light Conversion Screen (White)	90-15042-00
Blue Light Conversion Screen (Blue)	90-15043-00
Screen Holder	91-15116-00
Orange Filter (590/50 nm)	EBR-500K
Red Filter (620/40 nm)	SYPRO-500
Green Filter (537/35 nm)	91-13840-00
Blue Filter (460/40 nm)	HB-500
AlphaView Stand Alone software	91-14425-00
Imaging Test Target	85-13532-00
Magnetic Stylus	85-14195-00
Digital Thermal Grayscale USB Printer	91-12999-00
Thermal Paper, High Gloss, High Contrast, 5 rolls	P-200H
Thermal Paper, High Contrast, 4 rolls	P-150H
Spares/Service Parts	
302 nm UV Replacement Bulb	75-14084-00
365 nm UV Replacement Bulb	75-14083-00
Fuse, 250 V, 6 A	011-597

Part Description	Part Number
Service Contracts	
FluorChem E Platinum Service Contract	S-FCE-PL
FluorChem E Gold Service Contract	S-FCE-GS
FluorChem E Silver Service Contract	S-FCE-SS
FluorChem E Upgrade to Platinum Service	S-FCE-UGPL
FluorChem E Upgrade to Gold Service	S-FCE-UGGS
Preventive Maintenance Field Service	S-PMFC-FS
Preventive Maintenance Depot Service	S-PMFC-DS

Troubleshooting

Please contact ProteinSimple Technical Support at (888) 607-9692 or support@proteinsimple.com for troubleshooting, service or support-related items.

Chapter 11:

Analysis in Digital Darkroom

Chapter Overview

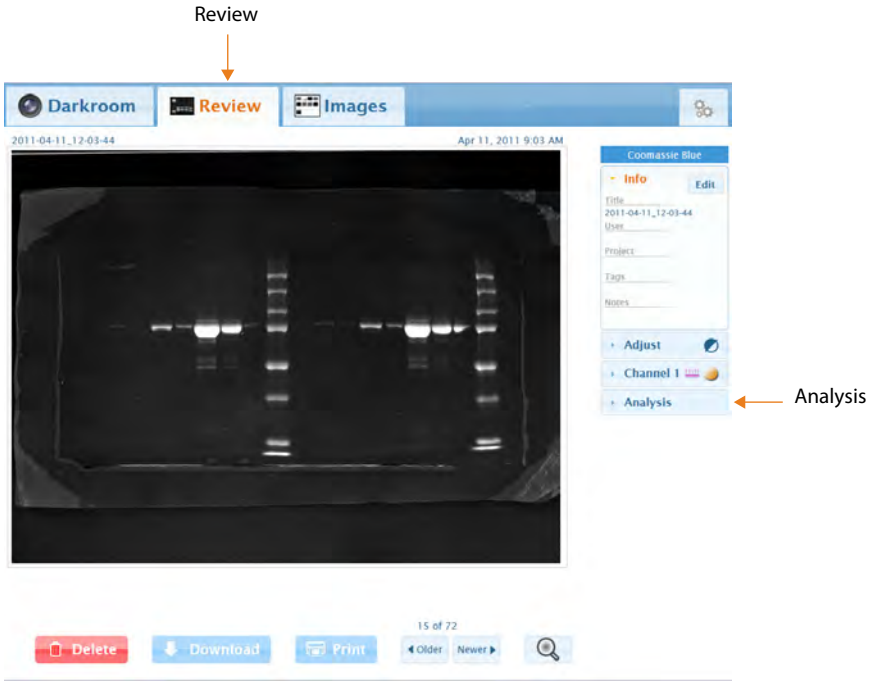
- Lane Profiling
- Summary Table
- Renaming Lanes
- Adding Molecular Weight Ladders
- Assigning Molecular Weight Ladders
- Exporting
- Printing

Lane Profiling

Lane profiling analysis is available through the Digital Darkroom software running on the FluorChem E. It can be used to measure densitometry or to determine molecular weights of samples.

Locating Analysis Tool

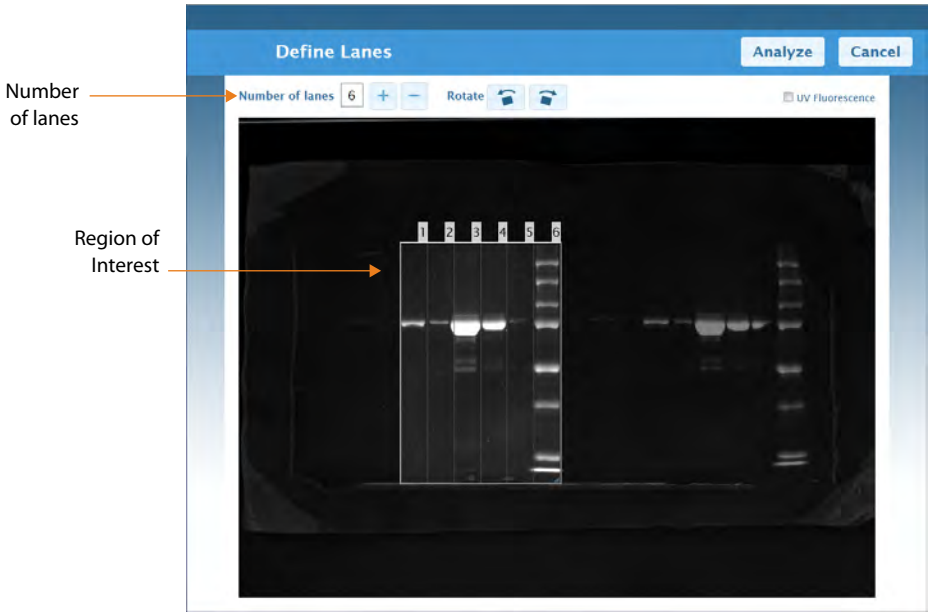
When an image is selected for **Review**, the **Analysis** tool becomes available. Click on the **Analysis** button to begin.



Defining Number of Lanes

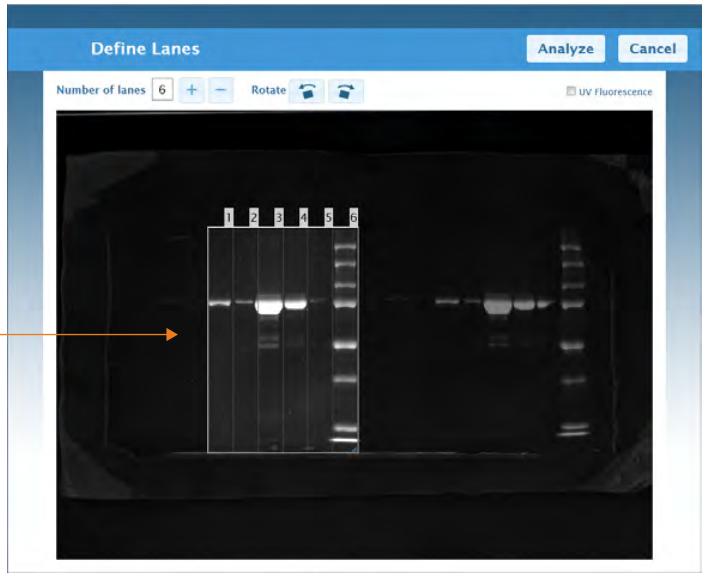
Clicking on **Analysis** expands the window. The next step will be to define your lanes. To do this, click on **Define Lanes**.

The number of sample lanes can be defined by entering the number of lanes into the text box or by using the **+** and **-** buttons to increase or decrease the number of lanes within the region of interest.



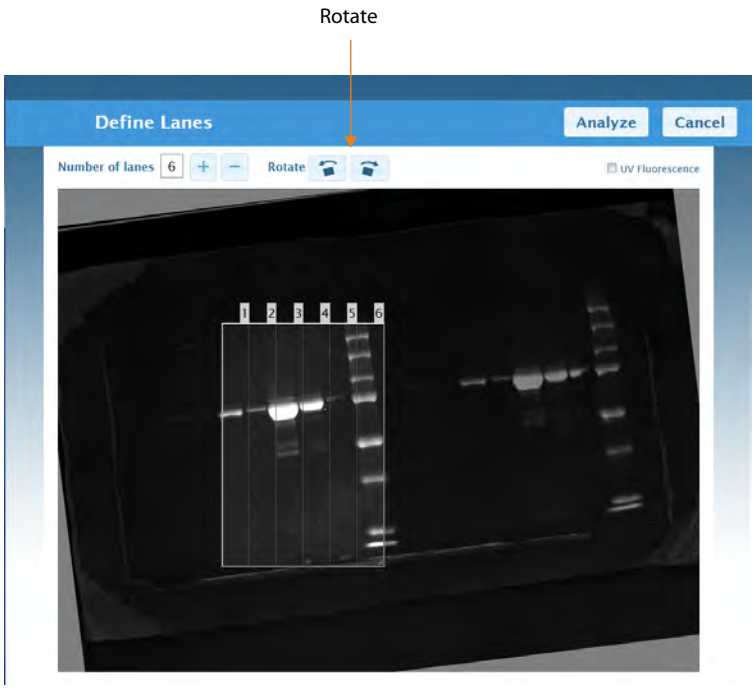
Defining Region of Interest

Define the region of interest by moving and resizing the rectangle overlaid on the gel or blot image. Select **Analyze** to continue or **Cancel** to leave analysis.



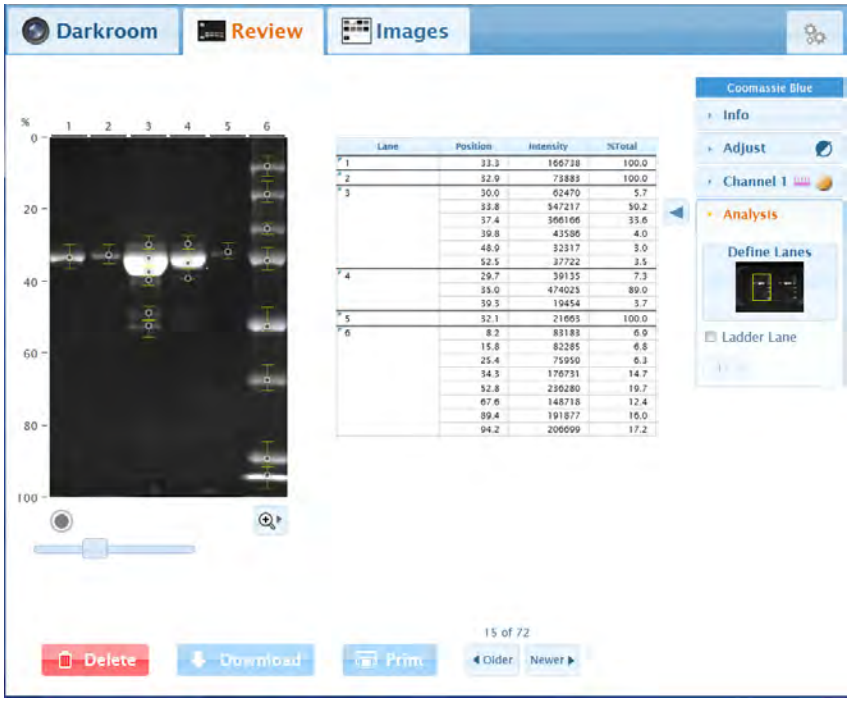
Rotating Region of Interest

The region of interest can be adjusted for rotation using the two **Rotate** buttons. Clicking on these buttons will rotate the image 1° in a clockwise or counterclockwise direction, with the center of rotation defined by the rectangular overlay.



Calculating Intensity Values

Based on the region of interest and number of bands, Digital Darkroom will automatically crop and zoom in on the area of interest, locate the bands in each lane, and calculate the intensity.



Band Selection

Individual bands can be selected and de-selected by double clicking at the desired position on the image.

Bands can be added or removed from the analysis using the threshold slider or by clicking on individual bands.

For a multi-channel image, use the round channel buttons to limit analysis to a single channel and to select the bands to be analyzed. To hide all the band indicators on the image, select a channel and toggle the bands off. Select **All** to analyze all the channels.

Move the threshold slider to raise or lower the intensity threshold for included bands.

The bands included are updated on the image as the slider is moved, the table updates after the slider is released.

Adding and Removing Individual Bands

To add a band to analysis:

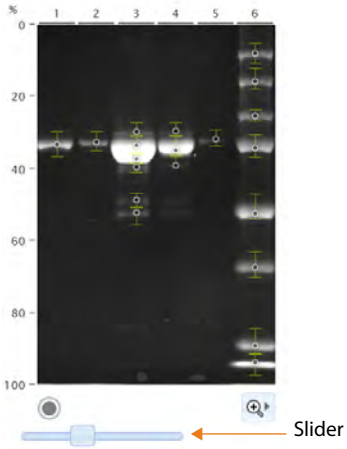
1. Move over the band on the image until the band indicator is shown.
2. Click on the band.

To remove a band from analysis:

1. Click on the band to highlight it in the table.
2. Click again to remove the band.

Band Sensitivity

The sensitivity of the software in detecting bands can also be set using the **Slider** bar.



Summary Table

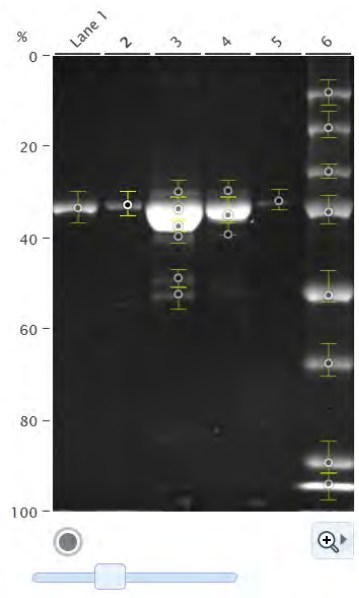
The summary table includes the **Lane** number, band **Position**, signal **Intensity**, and **%Total** each band contributes to the intensity of a lane. For multi-color images, reads from each wavelength will be color-coded.

- **Lane** - By default, the lanes are numbered starting with '1' on the left. The lane title can be edited in the table and will be displayed above the lane in the image.
- **Position** - The position if each band is reported as a percentage of the height of the selected region. Selecting a ladder lane will change the **Position** column to a molecular weight (**MW**) column.
- **Intensity** - Represents the total band intensity.
- **%Total** - Contribution of each band to the total channel intensity for the lane. The sum of all the bands in each channel is 100% within the rounding error.

Lane	Position	Intensity	%Total
1	33.3	166738	100.0
2	32.9	73883	100.0
3	30.0	62470	5.7
	33.8	547217	50.2
	37.4	366166	33.6
	39.8	43586	4.0
	48.9	32317	3.0
	52.5	37722	3.5
4	29.7	39135	7.3
	35.0	474025	89.0
	39.3	19454	3.7
5	32.1	21663	100.0
6	8.2	83183	6.9
	15.8	82285	6.8
	25.4	75950	6.3
	34.3	176731	14.7
	52.8	236280	19.7
	67.6	148718	12.4
	89.4	191877	16.0
	94.2	206699	17.2

Renaming Lanes

Lanes can be renamed by selecting the lane number in the table and typing into the text box.



Lane	Position	Intensity	%Total
Lane 1	33.3	166738	100.0
Lane 2	32.9	73883	100.0
3	30.0	62470	5.7
	33.8	547217	50.2
	37.4	366166	33.6
	39.8	43586	4.0
	48.9	32317	3.0
	52.5	37722	3.5
4	29.7	39135	7.3
	35.0	474025	89.0
	39.3	19454	3.7
5	32.1	21663	100.0
6	8.2	83183	6.9
	15.8	82285	6.8
	25.4	75950	6.3
	34.3	176731	14.7
	52.8	236280	19.7
	67.6	148718	12.4
	89.4	191877	16.0
	94.2	206699	17.2

Select Lane number

Adding Molecular Weight Ladders

To add a molecular weight ladder, check the box next to **Ladder Lane** in the **Analysis** section. Select the lane number your ladder is located in from the drop down menu.

Assigning Molecular Weight Ladders

Once a ladder lane has been selected, the molecular weight can be assigned by entering the molecular weight values directly into the table. To do this, click on the cell under **MW** that corresponds to the correct molecular weight and type. After the values are entered, the remainder of the table will automatically update.

Lane	MW	Intensity	%Total
Lane 1	36	166738	100.0
Lane 2	39	73883	100.0
3	58	62470	5.7
	33	547217	50.2
	9	366166	33.6
	-6	43586	4.0
	-66	32317	3.0
	-89	37722	3.5
4	59	39135	7.3
	25	474025	89.0
	-3	19454	3.7
5	44	21663	100.0
6	**200**	83183	6.9
	150	82285	6.8
	100	75950	6.3
		176731	14.7
		236280	19.7
		148718	12.4
		191877	16.0
		206699	17.2

Exporting

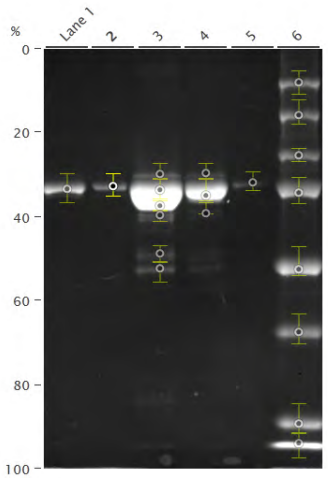
The analyzed image will reflect what is onscreen. Band identifiers may be hidden or only shown for a single channel. The tick marks may be percent or MW if entered in the table.

From the image **Review**, results of the analysis can be exported to a computer in two ways.

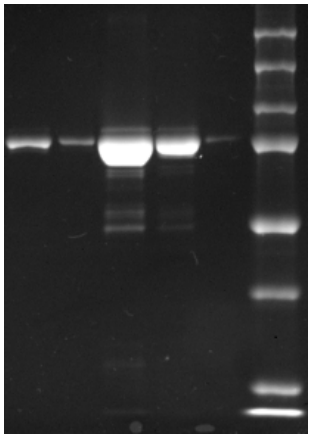
When exported as **Images (png files)**, there will be two additional files included in the zipped folder that will have the following added to the file name:

- _analyzed
- _lanes

The **_analyzed** .png will consist of the image cropped to the region of interest and any identified bands and named lanes.



The **_lanes** .png will consist only of the cropped image.



Printing

The image printed will always reflect what is onscreen. If analysis is enabled, the printed image will be cropped to the region of interest. To print the whole image, analysis must be hidden.